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## Phylogenomics of Andropogoneae (Panicoideae: Poaceae) of Mainland Southeast Asia

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**Abstract**—The grass tribe Andropogoneae is distributed in warm regions around the globe but has been poorly studied in mainland Southeast Asia. This is particularly true for the cosmopolitan genera *Andropogon* and *Schizachyrium*, with several species that appear to be narrowly distributed in this region. Additionally, lesser-known species in the genera *Hemisorghum*, *Kerriochloa*, and *Pseudosorghum* also occur in mainland Southeast Asia. A phylogeny is needed to address questions of taxonomy and trait evolution. Whole chloroplast genomes of Andropogoneae species and two outgroup species of *Garnotia* (tribe Arundinelleae) were analyzed using maximum likelihood (ML) and Bayesian inference (BI). Ancestral character states were reconstructed using ML for four morphological characters key to *Andropogon* and *Schizachyrium* identification. A previously-unidentified clade of Southeast Asian endemic taxa is found, including one species formerly classified in *Andropogon*. Other Southeast Asian taxa fall in an unresolved grade outside the major radiation of the tribe. *Andropogon* and *Schizachyrium* are both polyphyletic. Convergent evolution and reversal of characters are common throughout Andropogoneae. Addition of species from mainland Southeast Asian finds unexpected phylogenetic diversity. Southeast Asian *Schizachyrium sanguineum* forms two separate clades, which could reflect cryptic species differentiation, hybridization, introgression, or some combination.

**Keywords**—*Andropogon*, convergent evolution, *Schizachyrium*, Thailand, whole chloroplast genome.

Poaceae (the grass family) is one of the most diverse plant families in the world, containing approximately 12,000 species, many of which are ecologically and economically important (Clayton and Renvoize 1986; Watson and Dallwitz 1992; Kellogg 2015; Soreng et al. 2015). This is particularly true in the tribe Andropogoneae, which includes maize (*Zea mays* L.), sugarcane (*Saccharum officinarum* L.), sorghum (*Sorghum bicolor* (L.) Moench), Job's tears (*Coix lacryma-jobi* L.), and also ecologically dominant genera such as *Andropogon* L., *Arthraxon* P. Beauv., *Heteropogon* Pers., *Schizachyrium* Nees, and *Themeda* Forssk.

Andropogoneae is clearly monophyletic, and includes over 1,200 species in ca. 90 genera that dominate tropical and subtropical regions throughout the world (Clayton and Renvoize 1986; GPWG 2001; Chen et al. 2006; Simon 2007; Kellogg 2015). One putative synapomorphy for the tribe is the presence of paired spikelets, with one member of the pair sessile and the other pedicellate (Kellogg 2015). The phylogeny of the tribe has been difficult to resolve, apparently because of a rapid radiation early in the evolution of the tribe (Mathews et al. 2002; Spangler et al. 1999; Bouchenak-Khelladi et al. 2008). However, by using multiple concatenated nuclear genes, Estep et al. (2014) were able to identify early-diverging lineages including *Arthraxon*, *Chrysopogon* Trin., and a *Zea-Tripsacum* clade, a somewhat miscellaneous grade of smaller groups, and a *Saccharum* L. clade. About half of the species in the tribe were placed in a large well-supported clade that they called informally “the core Andropogoneae.” The “core” was made up of two subclades, one containing *Themeda*, *Heteropogon*, *Bothriochloa* Kuntze, *Dichanthium* Willemet, and *Capillipedium* Stapf, and the other containing *Diheteropogon* (Hack.) Stapf, *Andropogon*, *Schizachyrium*, and *Hyparrhenia* Andersson ex Fourn. (DASH clade).

Elements of the tribe from mainland Southeast Asia include *Dimeria* R. Br., *Eremochloa* Buse, *Germainia* Balansa & Poitr., *Hemisorghum* C.E. Hubb. ex Bor, *Kerriochloa* C.E. Hubb., and *Pseudosorghum* A. Camus (Clayton and Renvoize 1986; Buitenhuis and Veldkamp 2001; Neamsuvan et al. 2009; Teerawatananon et al. 2014). Few phylogenetic and evolutionary

studies have included these genera (but see Skendzic et al. 2007; Teerawatananon et al. 2011; Soreng et al. 2015). Consequently, additional data for these particular genera could help illuminate their evolutionary relationships, divergence pattern, and character evolution relative to other Andropogoneae species. Using species counts and data from floristic treatments, Hartley (1958) identified a center of diversity for Andropogoneae in Southeast Asia, and speculated that the tribe had originated in that part of the world, with subsequent radiations into drier areas such as Africa and parts of the western hemisphere.

Considering the Andropogoneae as a whole, two of the largest genera of the core are *Andropogon* and *Schizachyrium*, which together form a clade (Giussani et al. 2001; Mathews et al. 2002; Skendzic et al. 2007; Teerawatananon et al. 2011; Estep et al. 2014). These are major grasses for forage and ecosystems and have been used as representatives for many studies in ecology, taxonomy, and phylogenetics. *Andropogon*, the type genus of Andropogoneae, comprises over 100 species in tropical and subtropical regions of Africa, America, and Asia with a well-known diversity hotspot in Africa (Bor 1960; Anderson 1966; Gould 1967; Campbell and Windisch 1986; Clayton and Renvoize 1986; Zanin and Longhi-Wagner 2011; Nagahama and Norrmann 2012; Vorontsova et al. 2013). *Schizachyrium* includes ca. 60 species worldwide with an extensive distribution range in Africa, America, Asia, and Australia (Clayton 1972; Türpe 1984; Watson and Dallwitz 1992; Peichoto 2010).

*Andropogon* has no obvious synapomorphies. It is morphologically similar to *Schizachyrium*, from which it is distinguished by having paired or digitate branches in the inflorescence versus unbranched inflorescences in *Schizachyrium* (e.g. Clayton and Renvoize 1986). However, in a few species of *Andropogon* (e.g. *A. fastigiatus* Sw., segregated by some authors as *Diectomis fastigiata* (Sw.) P. Beauv.), the inflorescence branches are solitary as in *Schizachyrium*. In both genera, the sessile spikelet has bisexual florets while those of the pedicellate spikelet are either male or barren (Clayton and Renvoize 1986; Shouliang and Phillips 2006). Other

characters of the two genera are largely overlapping (Clayton et al. 2006).

Consistent with the lack of defining morphological characters, species of *Andropogon* and *Schizachyrium* often appear as intermixed in gene trees, and nuclear and chloroplast gene trees show unresolved and/or incongruent phylogenetic relationships. Depending on the taxon sample and marker used, *Andropogon* and *Schizachyrium* have appeared as polyphyletic or paraphyletic. No study has sampled the genera in any depth in any part of their range (Mathews et al. 2002; Skendzic et al. 2007; Teerawatananon et al. 2011).

In Thailand, species assigned to *Andropogon* and *Schizachyrium* occur in open areas of savanna, dry dipterocarp forest, and highland pine forest and are distributed throughout the country except in the south (Nanakorn 1990). Five species of *Andropogon* (*A. burmanicus* Bor, *A. chinensis* (Nees) Merr., *A. distachyos* L., *A. fastigiatus*, and *A. polyptychos* Steud.) and three species of *Schizachyrium* (*S. brevifolium* (Sw.) Nees ex Buse, *S. exile* (Hochst.) Pilg., and *S. sanguineum* (Retz.) Alston) are reported in Thailand. The morphological features of the species are largely distinct (Table 1), but, given questions about the limits of *Andropogon* in particular, it is unclear whether the Thai species are correctly assigned to genera. *Andropogon burmanicus* was mentioned in passing by Bor (1960), and is reported to have a narrow distribution, collected only from Myanmar before our accession was collected in Thailand (Bor 1960; Nanakorn 1990). The deeply-grooved lower glume of the sessile spikelet, rachis internode that extends into two lobes at the apex, and narrow distribution are distinctive for this species. *Andropogon distachyos* is geographically isolated, raising a question of whether it has been properly identified, classified, or is possibly introduced.

Advances in high throughput DNA sequencing have made recovery of chloroplast gene sets, and whole chloroplast genomes, increasingly easier (Steele et al. 2012; Stull et al. 2013). A number of studies have taken advantage of this technology, using either chloroplast protein-coding genes (e.g. Givnish et al. 2015; Washburn et al. 2015; Barrett et al. 2016) or complete chloroplast genomes (e.g. Carbonell-Caballero et al. 2015;

Cotton et al. 2015; Burke et al. 2016; McKain et al. 2016b) to resolve phylogenetic relationships from as deep as across Viridiplantae (Ruhfel et al. 2014) to within a species complex (e.g. *Mimulus* L.; Vallejo-Marín et al. 2016). The versatility of whole chloroplast genomes to resolve intrageneric relationships is promising for establishing evolutionary relationships in Andropogoneae of mainland Southeast Asia, an intratribal taxonomic issue spanning 26 million years (Estep et al. 2014).

In summary, this paper addresses the placement of mainland Southeast Asian species of *Andropogon* and *Schizachyrium* relative to each other and to other Southeast Asian Andropogoneae using sequences of whole chloroplast genomes (plastomes). We then investigate the evolution of taxonomic characters that distinguish the genera *Andropogon* and *Schizachyrium* from both each other and other members of Andropogoneae.

## MATERIALS AND METHODS

**Plant Materials**—Fifty-two species in 32 genera were sampled for this study. Multiple samples were included for many species, particularly those from mainland Southeast Asia in order to confirm the placement of the species, and to test for cryptic differentiation among populations. Of these specimens, 34 species of Andropogoneae and two species of Arundinelleae were collected from different locations in Thailand from September to December 2014 and leaf samples preserved in silica gel. In addition, 16 species of Andropogoneae were added from African and South American collections. For the latter, DNA was either extracted from leaves of field samples preserved in silica or from fresh leaves of samples grown from seed. All samples are listed in Appendix 1 with collector's numbers and herbarium locations. The geographic distribution of taxa is shown in Appendix 2.

**DNA Extraction, DNA Shearing, Library Preparation and Quantification**—DNA extractions were performed using a modified CTAB method (Doyle and Doyle 1987, 1990) for 0.5 mg of silica-dried plant leaves in each sample. DNA was run on a 1% agarose gel at 36 V for 1 h and 30 min to verify the quality and size distribution for library preparation. After DNA extraction, the concentration of DNA was adjusted for shearing and 50  $\mu$ l of each DNA sample was sheared with a Covaris S2 focused ultrasonicator targeting a distribution of fragments with a median of 500 bp. Whole genomic Illumina libraries were prepared using NEBNext ultra DNA library prep kit for Illumina (New England Biolabs, Ipswich, Massachusetts) using the manufacturer's protocol with the following

TABLE 1. Morphological comparison among *Andropogon* and *Schizachyrium* species in Thailand. SS, sessile spikelet; PS, pedicellate spikelet; glume 1, lower glume.

| Species                                   | Height (cm) | Leaf shape    | Leaf apex | SS to PS size | Primary infl. branches | Glume 1 of SS (abaxial side) | Wing on glume 1 of SS | Pedicel shape              | Glume 1 of PS | Apex of glume 1 of PS   | Longitudinal groove on pedicel | Rachis internode shape     |
|---|-------------|---------------|-----------|---------------|------------------------|------------------------------|-----------------------|----------------------------|---------------|-------------------------|--------------------------------|----------------------------|
| <i>A. burmanicus</i>                      | 100         | Linear        | Acuminate | SS = PS       | 2–3                    | Grooved                      | Absent                | Cuneate                    | Developed     | Muticous (Non-aristate) | Present                        | Cuneate                    |
| <i>A. chinensis</i>                       | 60–200      | Linear        | Acuminate | SS = PS       | 1                      | Convex                       | Absent                | Cuneate                    | Developed     | Aristate                | Absent                         | Cuneate                    |
| <i>A. distachyos</i> (type of the genus)  | 25–100      | Linear        | Acuminate | SS = PS       | 1                      | Convex                       | Present               | Linear to slightly cuneate | Developed     | Aristate                | Absent                         | Linear to slightly cuneate |
| <i>A. fastigiatus</i>                     | 15–200      | Linear        | Acuminate | SS < PS       | 0                      | Convex                       | Absent                | Cuneate                    | Developed     | Aristate                | Absent                         | Cuneate                    |
| <i>A. polyptychos</i>                     | 30–60       | Linear        | Acuminate | SS = PS       | 1–4                    | Convex                       | Absent                | Linear to slightly cuneate | Developed     | Muticous (Non-aristate) | Absent                         | Linear to slightly cuneate |
| <i>S. brevifolium</i> (type of the genus) | Up to 75    | Linear-oblong | Acute     | SS > PS       | 0                      | Convex                       | Absent                | Cuneate                    | Reduced       | Aristate                | Absent                         | Cuneate                    |
| <i>S. exile</i>                           | 30–50       | Linear-oblong | Acute     | SS > PS       | 0                      | Convex                       | Absent                | Cuneate                    | Reduced       | Aristate                | Absent                         | Cuneate                    |
| <i>S. sanguineum</i>                      | 80–120      | Linear        | Acuminate | SS > PS       | 0                      | Convex                       | Absent                | Cuneate                    | Developed     | Aristate                | Absent                         | Cuneate                    |

alterations. Libraries were size selected for a size of 600–800 bp using Agencourt AMPure XP beads (Beckman Coulter, Brea, California) as per the NEBNext ultra DNA library prep kit protocol. A total of 10 cycles was used for library amplification and final libraries were cleaned using Agencourt AMPure XP beads using a total volume of 0.9X beads and the NEBNext protocol. Samples were eluted in a final volume of 30  $\mu$ l nuclease-free water.

Library quality was determined by running samples on a 1% agarose gel at 36 V for 1 h and 30 min to identify size and absence of adapter dimers or over-amplification. A Qubit 2.0 fluorometer (Invitrogen Life Technologies, Carlsbad, California) was used to quantify double-stranded DNA concentrations (ng/ $\mu$ l). Molarity of libraries was calculated from an estimated average library length and concentration and adjusted to 10 nM for final library pooling. All libraries were pooled and the concentration adjusted to 10 nM for paired-end Illumina sequencing. Libraries were sequenced using a 2x150 Rapid Run on an Illumina HiSeq2500 at the New York University School of Medicine Genome Technology Center.

**Chloroplast Assembly and Annotation**—Prior to sequence assembly, sequence trimming and quality filtering of the reads were performed by Trimmomatic v.0.32 (Bolger et al. 2014) using the ILLUMINACLIP parameter for adapter trimming (TruSeq3-PE-2.fa:1:30:10), a sliding window of 10 base pairs with a minimum average quality score of 20 (SLIDINGWINDOW:10:20), and a minimum length of 40 bp (MINLEN:40). Filtered reads were then mapped to reference Andropogoneae plastomes using bowtie2 v.2.2.9 under the “very-sensitive-local” parameter set (Langmead and Salzberg 2012). Plastomes were initially assembled from mapped reads using SPAdes version 3.1.0 (Nurk et al. 2013) under the “only-assembler” option with k-mer sizes of 55, 87, and 121. SPAdes contigs were further assembled using the custom assembler afin (McKain et al. unpublished; <https://github.com/mrmckain/Fast-Plast>) with an initial contig trimming of 100 bp, an extension length of 100 bp, a stop extension minimum of 0.1, and 100 search loops to meta-assemble and fuse existing contigs. The genome of *Schizachyrium scoparium* (Michx.) Nash was fully assembled first, and all subsequent afin contigs were mapped to it as a reference to illustrate gene arrangement and provide scaffolding in partially complete plastomes. Coverage levels of afin contigs were estimated using the coverage analysis pipeline available in Fast-Plast (<https://github.com/mrmckain/Fast-Plast>). Cleaned reads were mapped to the contigs using bowtie2 v.2.2.9, using the “very-sensitive-local” parameter set. Jellyfish v.2.2.3 (Marçais and Kingsford 2011) was then used to estimate 25-mer abundance for mapped reads. K-mer abundance was then mapped across the contig sequences using a sliding 25 bp window. Boundaries between the large single copy (LSC), small single copy (SSC), and inverted repeats (IR) were initially identified using coverage, with the IR being approximately 2  $\times$  that of the single copy regions, and verified by sequence identity. Identified chloroplast regions were rearranged in the order LSC, IRB, and SSC and extended with a reverse complemented copy of IRB to represent the IRA segment in Geneious R8 (Kearse et al. 2012). For incomplete afin assembly files, contigs were assembled by in silico plastome-walking using cleaned reads to generate larger contigs until complete contigs were produced. In 18 out of a total of 76 plastomes, contigs of a plastome could not be joined, and N’s were inserted in the sequence equal to the distance between the scaffolded contigs as estimated by the alignment to *Schizachyrium scoparium*. Coverage analyses were conducted with all finished plastome assemblies before annotation and phylogenetic analysis as above. An assembly was considered acceptable when the plastome had greater than zero coverage for all non-N sequence windows less than 25 bp in length. All plastomes were annotated by annoBTD as implemented in Verdant (McKain et al. 2016a; <http://verdant.iplantcollaborative.org>). Final plastome sequences were deposited in GenBank (numbers in Appendix 1).

**Phylogenetic Analyses**—All analytical procedures were performed in the CIPRES Science Gateway 3.3 (Miller et al. 2010). The LSC, SSC, and IR were extracted from the whole plastome sequence using the `plastome_info_regions.pl` script (<https://github.com/mrmckain/Chloroplast-Phylogenomics-Utilities>) and separately aligned with MAFFT v.5 (Katoh et al. 2005) on XSEDE (Katoh et al. 2005). Aligned regions were concatenated into whole plastomes using the script `concat_plastomes.pl` (<https://github.com/mrmckain/Chloroplast-Phylogenomics-Utilities>). Arundinelleae, represented here by *Garnotia* Brongn., was selected as the outgroup for the ML reconstruction and BI, based on previous morphological and molecular data (Clayton and Renvoize 1986; Kellogg 2000; Teerawatananon et al. 2011; Soreng et al. 2015; Veldkamp et al. 2015). ML analysis was performed on the concatenated sequences under the General Time Reversible (GTR) model with the proportion of invariant sites and a gamma-shaped distribution of rates across sites (GTR +  $\Gamma$  + I) in RAXML v8.2.4

(Stamatakis 2006) with 1,000 bootstraps. For BI, MrBayes v3.2.6 was run (Ronquist et al. 2012) with the GTR + G + I model using four gamma rate categories and six substitution types. The Markov Chain Monte Carlo (MCMC) method was applied with two independent runs each time with four chains for a total of 10,000,000 generations each. FigTree v1.4.2 (Rambaut 2014) was used to visualize and edit for font type of taxa name and line thickening in the phylogenetic trees. Alignments have been deposited at Dryad (Arthan et al. 2017).

**Morphological Character Evolution**—We used the ML chloroplast phylogeny to investigate the evolution of inflorescence architecture, the extent of development of the pedicellate spikelet, and the relative sizes of sessile and pedicellate spikelets, characters that vary in and are used to distinguish the species investigated here (Table 1; Appendix 3). Data were based on descriptions in Clayton and Renvoize (1986), Watson and Dallwitz (1992), and Clayton et al. (2006), and supplemented and checked by our own observations. The pedicellate spikelet was scored as present, absent, or rudimentary. This character sometimes varies within a genus, so data presented here could be over-simplified particularly when only one species is sampled. Relative size of sessile and pedicellate spikelets was scored as having the sessile spikelet larger than, smaller than, or equal to the pedicellate one. A fourth state accommodated plants in *Dimeria ornithopoda* in which the sessile spikelet is absent.

Information on inflorescence architecture followed the morphological interpretations in Kellogg (2015). Specifically, Clayton and Renvoize (1986) use the terms “raceme” and “panicle” to describe grass inflorescences, but these terms are borrowed from dicots and fail to capture grass diversity. Instead we use two characters, number of primary branches and number of orders of branching. We consider primary branches to be those produced by the inflorescence meristem (IM) and bearing spikelet pairs, structures that Clayton and Renvoize (1986) call “racemes.” Barkworth (2003) calls these “rames” if they are disarticulating. While the spikelet pairs themselves can be considered primary branches if produced by the inflorescence meristem, they are developmentally and genetically different from long branches (Thompson 2014); spikelet pairs never become long branches in the course of normal development. The number of primary branches may be zero, with the IM producing only spikelet pairs (as in *Schizachyrium*, *Heteropogon*, and *Themeda*), fixed at one (i.e. the main axis and a branch, as in *Hyparrhenia* and *Cymbopogon* Spreng.), variable but rarely more than five (as in *Andropogon burmanicus*), and five to many. The latter state includes both species in which the primary branches are unbranched, such as *Arthraxon*, and those in which the branches go on to produce higher orders of branching, such as *Chrysopogon* and *Sorghum* Moench. For *Zea*, which has different branching patterns in the staminate and pistillate inflorescences (tassel and ear, respectively), we followed convention in the literature (e.g. Watson and Dallwitz 1992) and scored the morphology of the pistillate inflorescence.

In order to capture the distinction between the *Arthraxon*-type inflorescence and the *Chrysopogon-Sorghum* type, we use the character “number of orders of branching,” which reflects the fate of the primary branch meristem. In species with an unbranched axis, no primary branches are produced and thus the order of branching cannot be determined (coded NA). Species that produce only primary branches have a single order of branching (state 1). For species in which the primary branches themselves branch, the number of orders of branching is two or more (state 2). The latter state reflects inflorescences called “panicles” by Clayton and Renvoize (1986) and Clayton et al. (2006).

We did not attempt to assess whether the primary branches are digitate or sub-digitate, although this character appears commonly in keys. “Digitate or subdigitate branches” are in fact states of a quantitative character, “inflorescence internode length.” If internodes are very short, the branches appear digitate. Length of inflorescence internodes is controlled by different genes than the number of primary branches (the character illustrated) and is determined much later in the development (Doust et al. 2005; Kellogg et al. 2013). Primary branch number is determined soon after the transition of the shoot apical meristem from vegetative to inflorescence identity, whereas internode elongation occurs as the inflorescence is extending up through the leaf sheath. For all species with non-digitate branches (i.e. species with internode length more than about 1 mm), the character varies between internodes of an inflorescence and would need to be standardized, probably by internode position. It also varies within and between plants as well as between species and genera. Thus a meaningful analysis of the character would require extensive sampling and statistical analysis, neither of which was the goal of the present study.

Ancestral state reconstruction (ASR) for the above characters was conducted using a maximum likelihood method implemented in BayesTraits v.2.0 (Barker et al. 2007) on the plastome-based ML tree. The optimal

discrete character evolutionary model was identified using the *fitMK* function of Phytools v.5–10 (Lewis 2001; Revell 2012; Table 2). We compared the equal rates (“ER”) model, the symmetrical (“SYM”) model, and the all rates different (“ARD”) model. The ER model, as the name implies, has one rate of change for all states. The SYM model has variable rates for each state, but forward and reverse rates are equal. The ARD model has variable rates for all states in forward and reverse. Models of character evolution were tested using the Akaike information criterion (AIC) as implemented in *fitMK* for each trait. The ER model was found to be optimal for inflorescence branch number (delta AIC 4.84). The SYM model was optimal for the order of inflorescence branching (delta AIC 2.31) and relative size of the sessile spikelet to the pedicellate spikelet (delta AIC 7.16). The ARD model was found to be optimal for pedicellate spikelet presence (delta AIC 2.35). Trees were drawn using the *ape* v.3.4 (Paradis et al. 2004) module in R.

## RESULTS

**Chloroplast Genome Structure**—Chloroplast genome structure is largely conserved across all species in this study. Plastome sequence lengths range from at least 134,982 bp in *Heteropogon contortus* (L.) P. Beauv. ex Roem. & Schult. to 141,091 bp in *Eulaliopsis binata* (Retz.) C.E. Hubb., which are within the typical size range of grass plastomes (Palmer 1985; Hand et al. 2013; Cotton et al. 2015). Major divisions of the chloroplast genomes ranged in size from 80,172 to 83,733 bp for the LSC, 12,438 to 14,807 bp for the SSC, and 20,599 to 22,900 bp for the IR. Overall GC content was stable, ranging from 37.1 to 37.5%. IR boundaries are conserved for gene content in our sampled Andropogoneae with no gene movement in or out of the IR. *rps19* and *psbA* fall on either side of the LSC-IR<sub>A</sub> boundary, *rpl22* and *rps19* the LSC and IR<sub>B</sub> boundary, *rps15* and *ndhF* the SSC and IR<sub>B</sub> boundary, and *ndhH* and *rps15* the SSC and IR<sub>A</sub> boundary. Sampled chloroplast genomes demonstrate stability in overall architecture for Andropogoneae.

**Phylogenetic Analyses**—The LSC, SSC, and IR alignments were concatenated to produce a combined alignment of 133,506 characters. The tree topology was well resolved in ML analysis under the GTR+ $\Gamma$ +I model and had a likelihood (-lnL) of -335146.1879. BI produced a congruent topology (Fig. 1). High bootstrap values (BS > 90) and posterior probabilities (PP = 1) supported most nodes except for the one at the divergence of the *Kerriochloa* clade from the rest of Andropogoneae (BS = 67 and PP = 0.96), the *Mnesithea-Eremochloa* clade and the rest of Andropogoneae (BS = 77 and PP = 0.98), the *Heteropogon triticeus* clade and *Cymbopogon* (BS = 82 and PP = 0.98), and within the *Heteropogon triticeus* clade (BS = 81 and PP = 0.33), the *Andropogon chinensis* clade (BS = 71 and PP = 0.42), and the *Schizachyrium sanguineum* clades (BS = 81 and 64 and PP = 0.81 and 0.81, respectively).

Multiple accessions were sequenced for twelve species (*Andropogon burmanicus* (2 accessions), *A. chinensis* (3), *A. fastigiatus* (4), *Arthraxon lanceolatus* (Roxb.) Hochst. (2), *Eriochrysis cayennensis* P. Beauv. (2), *Heteropogon triticeus* (4), *Hyparrhenia rufa* (Nees) Stapf (2), *Kerriochloa siamensis* C.E.

Hubb. (2), *Polytoca digitata* (L. f.) Druce (2), *Pseudosorghum fasciculare* (Roxb.) A. Camus (2), *Schizachyrium brevifolium* (5), and *S. sanguineum* (6). These accessions form strongly supported species-specific clades, as indicated by the heavy lines in Fig. 1. The exception is *S. sanguineum*, in which we found two well-supported monophyletic sets of accessions (Fig. 1). One clade was sister to *S. scoparium* plus *S. spicatum* (Spreng.) Herter, and the other was grouped with *S. exile* with both supported by bootstrap values of 100 and posterior probabilities of 1.

Many genera are para- or polyphyletic. *Schizachyrium* forms a polyphyletic group with *S. imberbe* A. Camus and *S. tenerum* Nees forming a clade sister to *A. fastigiatus* with bootstrap value and posterior probability of 100 and 1, respectively; this *Schizachyrium*+*A. fastigiatus* clade is then sister to a clade comprised of the remaining *Schizachyrium* species and *A. chinensis* (Fig. 1). *Andropogon* is polyphyletic; *A. chinensis* is sister to most species of *Schizachyrium*, *A. fastigiatus* is sister to *S. imberbe* plus *S. tenerum*, *A. distachyos* and *A. abyssinicus* R. Br. ex Fresen. together are sister to *Hyparrhenia*, and *A. burmanicus* is sister to *Eulalia contorta* (Brongn.) Kuntze (Fig. 1). *Eulalia* Kunth is also polyphyletic and *Heteropogon* is paraphyletic.

After rooting the tree at *Garnotia* (Arundinelleae, or Andropogoneae s. l.), the *Arthraxon* clade (represented by *A. hispidus* (Thunb.) Makino, *A. lanceolatus*, *A. microphyllus* (Trin.) Hochst., and *A. prionodes* (Steud.) Dandy) was sister to the rest of the Andropogoneae with strong support (BP = 100 and PP = 1). The core Andropogoneae is made up of two major subclades: the *Andropogon-Schizachyrium-Hyparrhenia* clade (BS = 100 and PP = 1) and the *Themeda-Heteropogon-Cymbopogon-Dichanthium* clade (BS = 99 and PP = 1).

Outside the core is a set of smaller, less well-defined groups. Of particular interest here is the clade made up of *Eriochrysis* P. Beauv., *Chrysopogon*, *Andropogon burmanicus*, *Eulalia contorta* and *Parahyparrhenia siamensis* A. Camus. Many of these taxa are Southeast Asian and poorly known. *Coix* L. and *Kerriochloa* were isolated and their position was not resolved in the trees. *Mnesithea* Kunth and *Eremochloa* formed a monophyletic group. Also, a monophyletic clade was observed for *Eulaliopsis* Honda and *Dimeria*. *Germainia*, *Pogonatherum* P. Beauv., *Hemisorghum*, *Sorghum*, *Eulalia siamensis* Bor. and *Pseudosorghum* were grouped in a single clade (BS = 100 and PP = 1).

**Analysis of Character Evolution**—Inflorescence architecture is variable and dynamic among Andropogoneae (Fig. 2). For the number of primary branches (i.e. branches that are the product of the IM), the analysis favored a model in which all character state changes are equally likely. Given this taxon sample, having an unbranched inflorescence is most likely to be ancestral for the core Andropogoneae, and indeed for all of Andropogoneae excluding *Garnotia*, though there is much uncertainty in the backbone of the tree (Fig. 2A). Having exactly one primary branch (i.e. the main axis and one branch) is derived independently several times as are the states of having

TABLE 2. Morphological characters, character states, best fit model, and delta AIC scores. ER = equal rates; SYM = symmetrical rates; ARD = all rates different; SS = sessile spikelet; PS = pedicellate spikelet.

| Character                         | States                               | Best Model | delta AIC |
|-----------------------------------|--------------------------------------|------------|-----------|
| Primary inflorescence branches    | 0, 1, 2–4, 4+                        | ER         | 4.8351    |
| Orders of inflorescence branching | 0, 1, 2                              | SYM        | 2.3075    |
| Size of SS relative to PS         | SS > PS, SS < PS, SS = PS, SS absent | SYM        | 6.4507    |
| Pedicellate spikelet              | Present, absent, rudimentary         | ARD        | 2.3490    |

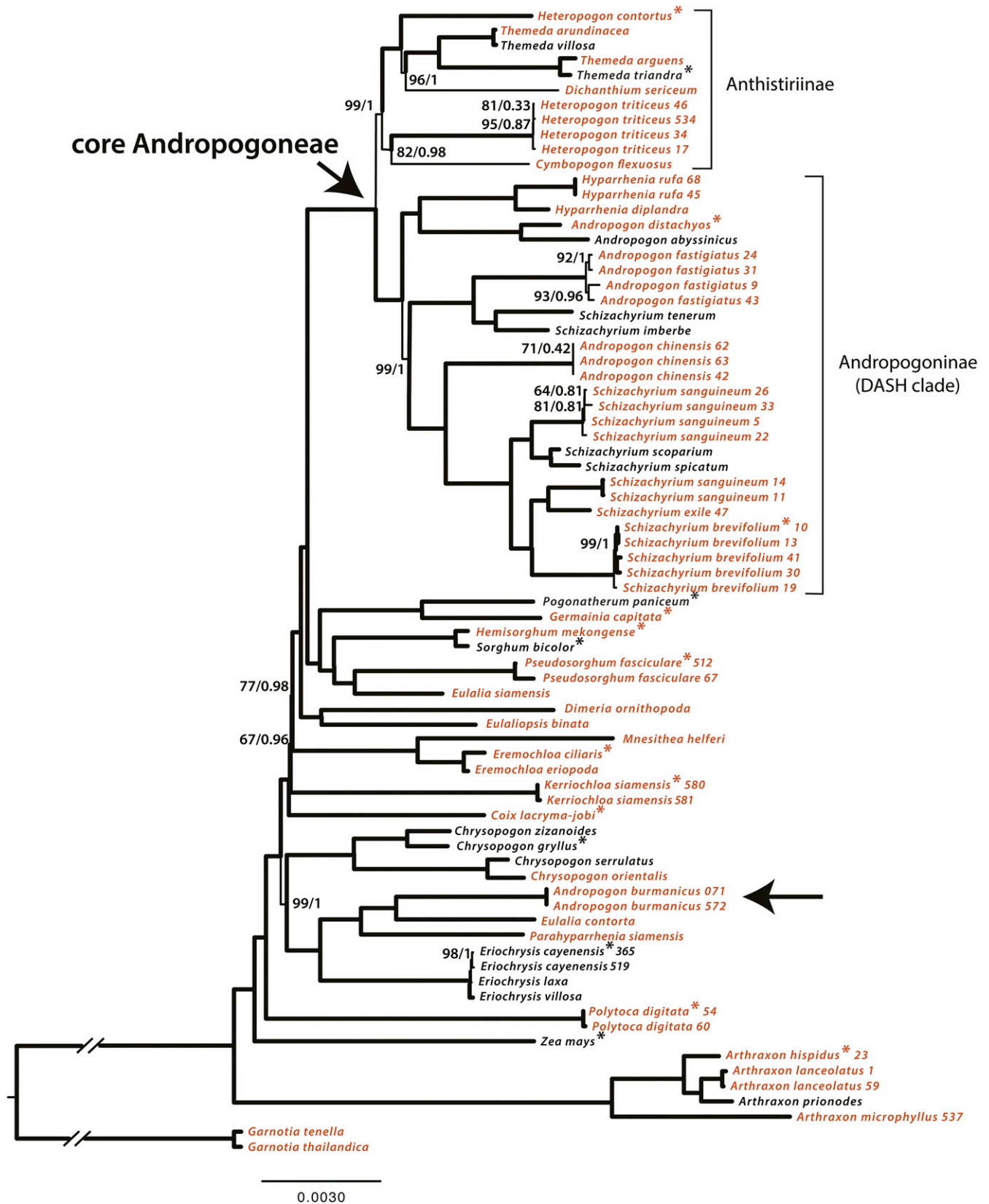


FIG. 1. Maximum likelihood tree based on whole chloroplast genome data of 76 Andropogoneae taxa and produced by RAxML under the general time reversible evolution model (GTR +  $\Gamma$  + I). Unlabeled nodes and heavy lines have ML bootstrap of 100 and posterior probability of 1. Nodes with lower values are labeled as ML bootstrap / posterior probability and indicated by lighter lines. Taxa from Southeast Asia are in orange. Species that are types for their respective genera are noted with an asterisk. Collection numbers are included only where there are multiple accessions of the same species. Core Andropogoneae is well-supported as are subtribes Andropogoninae and Anthistirinae. A horizontal arrow points to *Andropogon burmanicus*, which is unrelated to other species of *Andropogon*.



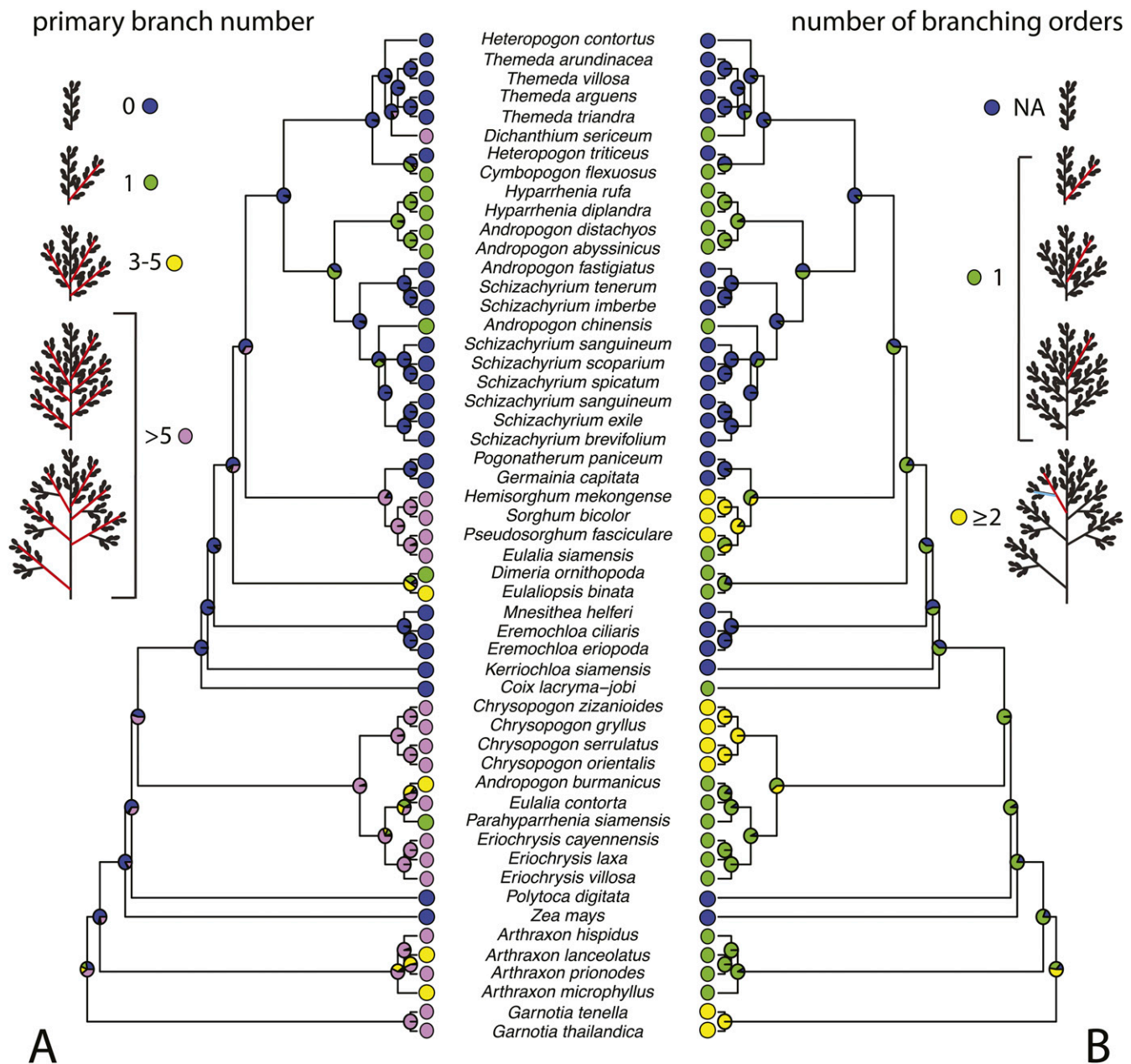


FIG. 2. Evolution of inflorescence branching traits. A. Number of primary branches, products of the inflorescence meristem. Excluding the spikelet pairs, the number of branches produced by the inflorescence meristem is either zero (blue circles), fixed at one (green), fixed at 3 to 5 (yellow), or greater than 5 (violet). Primary branches are shown in red. B. Number of orders of branching, products of the branch meristems. Excluding the spikelet pairs, the number of order of branching is either not applicable (blue circles, the inflorescence unbranched), one (green, primary branches do not themselves branch), or greater than or equal to two (yellow, higher order branches are present). Representative primary branches are shown in red, secondary branch in blue.

3 to 5 branches and having many branches (Fig. 2A). Number of orders of branching varies between zero (not applicable) and one throughout most of the tree with potentially three to four origins of second order branching in *Garnotia*, *Chrysopogon*, and the *Sorghum*+*Hemisorghum*+*Pseudosorghum*+*E. siamensis* clades (Fig. 2B); this model is symmetrical suggesting that reversals of states are as likely as forward changes. The ancestral number of orders of branching appears to contradict the optimization of the ancestral state of “unbranched” for numbers of axes. The interpretation of this is discussed below.

The ancestral state for pedicellate spikelet development is also completely ambiguous. The ARD (all rates different) model is the best fit of the models that we tested (Table 2).

Under this model, the probabilities of change from rudimentary to pedicellate and from absent to pedicellate are quite high (1,000 and 398, respectively), whereas the probability of change from pedicellate to rudimentary or pedicellate to absent is much lower, and the probability of change from rudimentary to absent is zero. If the ancestral state is for the pedicellate spikelet to be present (blue), then pedicellate spikelets could have been reduced or lost up to six times in the evolution of the tribe (Fig. 3A), in *Garnotia thailandica* Gould, *Arthraxon hispidus*, *Zea mays*, *Coix lacryma-jobi*, *Eremochloa*+*Mnesithea*, and one clade of *Schizachyrium*. However, the ancestral state could have been “absent,” in which case there could have been many origins of pedicellate spikelets.

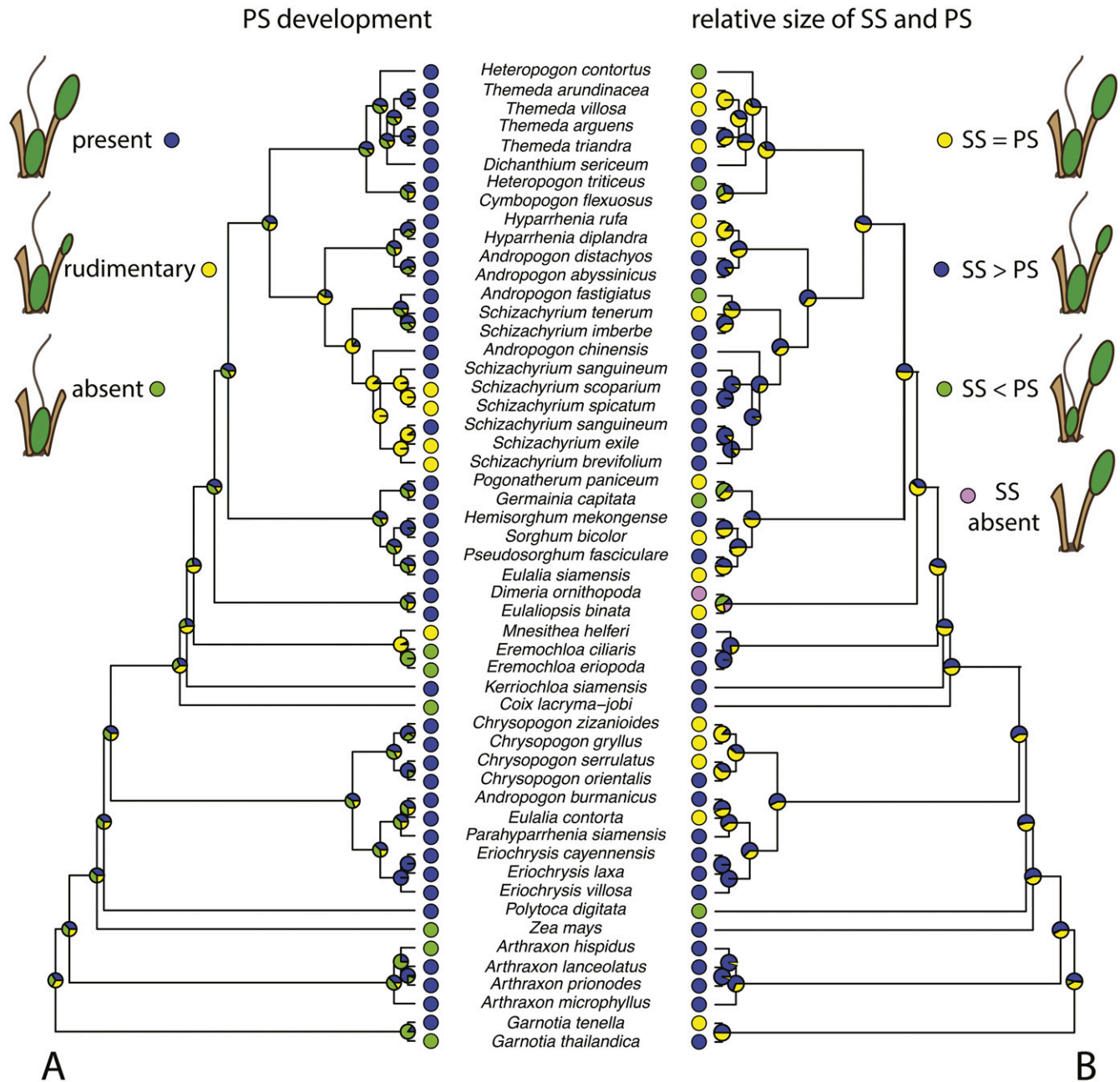


FIG. 3. Pedicellate spikelet (PS) development and relative size. A. the pedicellate spikelet may be present (blue circles), rudimentary (yellow), or absent (green). B. the sessile and pedicellate spikelets may be the same size (SS = PS, yellow), the sessile spikelet may be larger than the pedicellate spikelet (SS > PS, blue), the sessile spikelet may be smaller than the pedicellate spikelet (SS < PS, green), or the sessile spikelet may be absent (violet, only *Dimeria*). The cartoon of each sessile-pedicellate pair shows, from left to right, the attached inflorescence internode (brown), the sessile spikelet (green) with its attached awn, the pedicel (brown), and the pedicellate spikelet.

Relative size of the two spikelets of a pair (Fig. 3B) also varies between species and best fits a symmetrical model. Within the DASH clade in particular sessile spikelets may be smaller than, equal to, or larger than pedicellate spikelets. *Dimeria ornithopoda* Trin. is unique in our sample, as it does not have sessile spikelets at all. *Andropogon burmanicus* shares similar character states to other *Andropogon* species, as it has pedicellate spikelets that are smaller than the sessile spikelets. This character combination is shared with closely related species like *Parahyparrhenia siamensis* and the genus *Eriochrysis*. Ancestral state reconstructions point to the ancestral state being for the pedicellate spikelet to be the same size as or smaller

than the pedicellate one. Under this optimization, having a pedicellate spikelet larger than the sessile one has originated independently five times.

#### DISCUSSION

***Andropogon* and *Schizachyrium***—The original goal of this study was placement of the Thai species of *Andropogon* and *Schizachyrium*. Like most previous studies, we found a clade including *Andropogon*, *Schizachyrium*, and a monophyletic *Hyparrhenia* (Mathews et al. 2002; Skendzic et al. 2007; Estep et al. 2014). *Andropogon* and *Schizachyrium* are morphologically



similar and have always been assumed to be closely related (Clayton 1964; Clayton 1972; Estes and Tyrll 1976; Clayton and Renvoize 1986). Likewise, the monophyly of *Hyparrhenia* has never been questioned, with all species having exactly two inflorescence axes (the main axis and one primary branch) and having the callus of the sessile spikelet applied obliquely to the rachis (Clayton et al. 2006; Kellogg 2015). In contrast, *Andropogon* inflorescences vary in number of primary branches, and *Schizachyrium* has none; in both genera the callus is inserted into the cup-like apex of the internode below (Clayton et al. 2006; Kellogg 2015).

*Andropogon* is clearly polyphyletic, with species appearing even more widely scattered in the phylogeny than in previous studies (e.g. Estep et al. 2014), probably as a result of our sample of more disparate members of the genus. Although this study still includes only a fraction of the species in the genus, we think it is unlikely that inclusion of additional species will cause *Andropogon* to appear as monophyletic. A phylogeny including 40 species of *Andropogon* retrieves results consistent with those presented here (McKain and Kellogg, unpublished data). *Andropogon abyssinicus* and *A. distachyos* formed a clade sister to *Hyparrhenia* with which they share geographic provenance in Africa (Skendzic et al. 2007; Nagahama and Norrmann 2012) and an inflorescence with exactly two axes. *Andropogon distachyos* is rare in Thailand and might have been imported for fodder. Both *A. abyssinicus* and *A. distachyos* have a winged lower glume on the sessile spikelet, a character that is not reported for *Hyparrhenia*.

*Andropogon fastigiatus* is on a long branch sister to *Schizachyrium imberbe* and *S. tenerum*, which together have been placed in *Schizachyrium* Section A (Fig. 1; Clayton 1964). Like *Schizachyrium*, *A. fastigiatus* has an unbranched inflorescence (Fig. 2A, B). It is sometimes segregated as its own genus, *Diectomis* Kunth, based on its enlarged pedicellate spikelet, a character that is unusual in the *Andropogon-Schizachyrium-Hyparrhenia* clade (Fig. 3B). The *A. fastigiatus* clade includes two subclades indicating different types of plastomes in these populations (Fig. 1).

Placement of *A. burmanicus* in a clade with *Eulalia contorta* and *Parahyparrhenia siamensis* was unexpected but was confirmed by sequences from two plants collected at different times (Appendix 1). The plants appear morphologically similar to other species of *Andropogon* in having a small number of inflorescence axes which do not branch further (Fig. 2A, B; Bor 1960 and personal observations) in addition to a caespitose habit. Also like other *Andropogon* species, *A. burmanicus* has a well-developed pedicellate spikelet (Fig. 3A), although the pedicellate spikelet is smaller than the sessile one (Fig. 3B). It also has a deeply grooved lower glume on the sessile spikelet, a character that appears in some species of *Andropogon* as well as in other species throughout the tribe. In contrast to *A. burmanicus*, the inflorescence in *Eulalia contorta* has more branches, and that of *Parahyparrhenia siamensis* has only one in addition to the main axis (Fig. 2A). However, all three species have only first order branches (Fig. 2B). *Andropogon burmanicus* also has a pyriform and ciliate rachis internode and pedicel, and an appendaged pedicellate spikelet (Chen et al. 2012) similar to that found in some species of *Microstegium* Nees (e.g. *M. eucnemis* (Nees ex Steud.) A. Camus). However, *A. burmanicus* exhibits a pronounced longitudinal groove on the rachis internode, and leaf blades that are narrow and have a waxy abaxial surface creating a distinctive bicolored appearance; none of these characters are described in

*Microstegium*. Because *Microstegium* was not included in this analysis, more extensive sampling is required to determine the exact position of *A. burmanicus*.

*Schizachyrium* contains two separate lineages, one clade with the majority of *Schizachyrium* species and the other of *S. imberbe* and *S. tenerum* (Fig. 1). The latter two species were placed in *Schizachyrium* section A by Clayton (1964) based on their glabrous rachis internodes and pedicels, well-developed pedicellate spikelets (Fig. 3B), and shortly bilobed upper lemmas (Türpe 1984; Peichoto 2010), similar to their sister species *Andropogon fastigiatus*.

In contrast, the members of the larger clade, proposed as section B by Clayton (1964), share pubescent rachis internodes and pedicels, reduced pedicellate spikelets, and deeply bilobed upper lemmas. The trend raises the hypothesis that the surface of the rachis internode or pedicel, the development of the pedicellate spikelet, and the shape of the upper lemma could be synapomorphic for major groups in *Schizachyrium*. However, much more extensive taxon sampling, particularly from Africa and South America, is recommended for future study.

*Andropogon chinensis* is sister to *Schizachyrium* section B, from which it differs by having one long branch in addition to the main inflorescence axis (Fig. 2A) and well-developed pedicellate spikelets (Fig. 3A). However, it is similar in having pubescent rachis internodes and pedicels.

Two distinct groups of *S. sanguineum* were discovered in this study (Fig. 1), although their morphology is apparently indistinguishable. One group of *S. sanguineum* plastomes clustered with American *Schizachyrium* (*S. scoparium* and *S. spicatum*) and the other with Asian species (*S. exile* and *S. brevifolium*). All *S. sanguineum* samples were collected in Thailand, suggesting either a deep coalescence of chloroplast haplotypes within *S. sanguineum* that predate the geographical disjunction of other *Schizachyrium* species or admixture of haplotypes through allopolyploidy as discussed below (Peichoto et al. 2011; Estep et al. 2014).

**The Core Andropogoneae**—The core Andropogoneae has been widely accepted and retrieved in all phylogenetic studies to date, including analyses of nuclear genes such as PHYB (Mathews et al. 2002), GBSSI (Mason-Gamer et al. 1998; Mathews et al. 2002), single-copy genes (Estep et al. 2014; Welker et al. 2015, 2016), and ITS (Skendzic et al. 2007), chloroplast genes such as *ndhF* (Giussani et al. 2001; Mathews et al. 2002) and *trnL-F* (Teerawatananon et al. 2011), and combined data. Two clades are clearly found: 1) the group of *Cymbopogon*, *Dichanthium*, *Heteropogon*, and *Themeda* and 2) the group of *Andropogon*, *Hyparrhenia*, and *Schizachyrium*.

Two subclades based on inflorescence shape appear in *Themeda*, spindle-shaped in *T. arundinacea* (Roxb.) A. Camus and *T. villosa* (Poir.) A. Camus and fan-shaped in *T. triandra* Forssk. and *T. arguens* (L.) Hack., with the shape difference caused by the arrangement of the homogamous spikelets at different and equal levels, respectively. Interestingly, *Heteropogon* is paraphyletic. The two species of this genus included here are similar in having a long and bearded callus, awnless homogamous spikelets at the base of the raceme, and awned sessile spikelets on the top portion of the raceme. It will be interesting in the future to include the remaining four species of *Heteropogon*.

**Mainland Southeast Asia Contains Unexpected Phylogenetic Diversity**—This study discovered unexpected diversity among Andropogoneae of mainland Southeast Asia, implying

that the endemic grass flora of Southeast Asia is richer in Andropogoneae genera than previously thought. More than half a century ago, Hartley (1950, 1958) proposed the hypothesis that Southeast Asia might be the origin of the Andropogoneae. However, this interesting possibility needs to be optimized on a more completely sampled phylogeny to answer this question.

Based on overall morphological similarity (as described in, for example, Clayton 1972; Clayton and Renvoize 1986; Watson and Dallwitz 1992; Veldkamp 2003; Soreng et al. 2015), we had expected that *Andropogon burmanicus* would fall within *Andropogon*, *Parahyparrhenia siamensis* would be closely related to *Hyparrhenia*, and *Pseudosorghum fasciculare* would be close to *Sorghum*. Instead *A. burmanicus* and *Parahyparrhenia siamensis* are in a clade with *Eulalia contorta* that is entirely Southeast Asian, but sister to the widespread genus *Eriochrysis*. *Pseudosorghum fasciculare* is sister to *Eulalia siamensis*, and neither is closely related to *Eulalia contorta*, supporting the polyphyly of *Eulalia* as noted by other authors (Skendzic et al. 2007; Teerawatananon et al. 2011; GPWG II 2012; Estep et al. 2014; Soreng et al. 2015; Welker et al. 2016).

Likewise, Southeast Asian species such as *Polytoca digitata*, *Kerriochloa siamensis*, and *Mnesithea helferi* (Hook. f) de Koning & Sosef fall in clades outside the core Andropogoneae, and are not embedded in larger genera (Fig. 1). *Kerriochloa siamensis* is in an isolated position in this study as in the analyses of combined chloroplast genes and ITS data from Teerawatananon et al. (2011). Estep et al. (2014) placed it as the sister to *Microstegium vimineum* (Trin.) A. Camus and *Sehima nervosum* (Rottler ex Roem. & Schult.) Stapf, but neither of these species was included in the current analysis. The grouping of *Mnesithea* and *Eremochloa* is supported by the appearance of numerous appendages on the rim of the lower glume in *M. glandulosa* (Trin.) de Koning & Sosef, resembling those in *Eremochloa* (Buitenhuis and Veldkamp 2001; Traiperm 2007; Veldkamp et al. 2013). These two genera are morphologically similar to other members of subtribe Rottboelliinae in that they lack an awn on the upper lemma (Clayton and Renvoize 1986; Kellogg 2015; Soreng et al. 2015).

The Southeast Asian species *Eulaliopsis binata* falls in a clade with *Dimeria ornithopoda*, although the two share no obvious characteristics. The former species has multiple inflorescence branches, whereas the latter has only one in addition to the main axis (Fig. 2A). *Dimeria* is also the only member of Andropogoneae that lacks a sessile spikelet (Fig. 3B; Kellogg 2015).

*Germainia* and *Pogonatherum*, which are strongly supported as sisters, both have sessile spikelets with the upper floret male and the pedicellate spikelet female or bisexual, reversing the condition in most Andropogoneae (Clayton and Renvoize 1986; Clayton et al. 2006; Kellogg 2015). In other Andropogoneae, the awn occurs on the lemma of the seed-bearing floret, which is normally in the sessile spikelet. In *Germainia* and *Pogonatherum*, however, the lemma of the upper floret of the pedicellate spikelet is awned, indicating that presence or absence of the awn is correlated with development of a functional ovary.

*Hemisorghum*, *Pseudosorghum*, and *Sorghum* are phylogenetically related and are morphologically similar in having robust highly branched inflorescences and 2-keeled lower glumes (Clayton and Renvoize 1986; Neamsuvan et al. 2009). *Hemisorghum mekongense* (A. Camus) C.E. Hubb. was originally described as a species of *Sorghum* but was segregated

by Hubbard in Bor (1960). Our data show that it is closely related.

The position of *Eulalia siamensis* as sister to *Pseudosorghum fasciculare* is hard to explain morphologically. Like other species classified in *Eulalia* and unlike other members of its clade, the sessile and pedicellate spikelets are morphologically indistinguishable (Fig. 3B). In addition, *E. siamensis* exhibits only one order of branching, whereas the other members of the clade have highly branched inflorescences (Fig. 2B).

**Arthraxon, Polytoca, and Zea**—The placement of *Arthraxon*, *Zea*, and *Polytoca* R. Br. as successive sisters to the rest of Andropogoneae is consistent with the results of other studies (Giussani et al. 2001; GPWG II 2012; Soreng et al. 2015). Our data show that *Arthraxon* is monophyletic, in accord with other taxonomic and phylogenetic studies (Bouchenak-Khelladi et al. 2008; Christin et al. 2009; Teerawatananon et al. 2011; Estep et al. 2014; Kellogg 2015; Soreng et al. 2015; Welker et al. 2015). The species of *Arthraxon* display broad-lanceolate to ovate leaves clasping the culm, and have an awn originating at the base of the fertile upper lemma in the sessile spikelet (Clayton 1972; Van Welzen 1981; Teerawatananon et al. 2011). *Arthraxon hispidus* was found to have distinctive cells in the leaves, which are not commonplace in Andropogoneae but could be detected in the genera *Arundinella* Raddi and *Garnotia*, the sister genera to Andropogoneae (Ueno 1995). Thus the presence of distinctive cells might be a shared character in the common ancestor of Arundinelleae and Andropogoneae.

*Zea mays* diverges after the *Arthraxon* clade consistent with previous phylogenies of chloroplast genes (e.g. Bouchenak-Khelladi et al. 2008; Soreng et al. 2015). Geographically, the genus *Zea* originated in Mexico (Doebley 1990; Jannink and Veldkamp 2002; Zuloaga et al. 2007), while the center of diversity of *Arthraxon* is Africa and India (Van Welzen 1981).

We scored *Zea* as having an unbranched inflorescence (Figs. 2A, B), following the convention in the grass literature to report the morphology of the female-fertile spikelet and inflorescence (see Watson and Dallwitz 1992). However, the male inflorescences have many primary branches and thus are more similar in that character to *Arthraxon*. Many other characters differ between the male and female inflorescences of *Zea*. Because of its position near the base of the tree, the states of these characters could easily influence estimation of the ancestral state for the tribe. Likewise, we scored *Polytoca* as having an unbranched inflorescence, although *P. wallichiana* (not sampled here) may have many inflorescence branches. In both species, the structure of the staminate and pistillate parts of the inflorescence is somewhat different. Estep et al. (2014) demonstrated that there is considerable diversity found between *Zea* and the core Andropogoneae that is not sampled here. A more diverse set of genera within Andropogoneae is needed to resolve relationships and patterns of morphological evolution along the backbone of the phylogeny.

The phylogeny (Fig. 1) includes representatives of all four monoecious clades in the tribe. Other data (e.g. Estep et al. 2014) show that *Zea* is sister to the monoecious *Tripsacum*, *Polytoca* is sister to *Chionachne*, *Coix* is of uncertain placement, and *Heteropogon contortus* falls among the core taxa. Therefore, additional sampling is not likely to change our view of the origin of monoecy in the tribe.

**Chrysopogon and Eriochrysis**—Both *Chrysopogon* and *Eriochrysis* are monophyletic, consistent with previous results

(Mathews et al. 2002; Estep et al. 2014; Welker et al. 2016). *Chrysopogon* species fall into two groups, the clade of *C. gryllus* (L.) Trin. and *C. zizanioides* (L.) Roberty, and that of *C. orientalis* (Desv.) A. Camus and *C. serrulatus* Trin.

*Eriochrysis* forms a well-supported clade and is sister to a clade of endemic Southeast Asian species. Although all samples of *Eriochrysis* in the analyses are American, members of the genus *Eriochrysis* are also distributed in Africa and India (Clayton and Renvoize 1986; Kellogg 2015; Welker and Longhi-Wagner 2012; Welker et al. 2016). In future studies, African or Indian *Eriochrysis* species should be included to determine whether they are more closely related to the Southeast Asian ones and diverge before the American species as suggested by Welker et al. (2016).

**Implications for Classification**—The phylogeny presented here contains a broad sample of genera of Andropogoneae, but because of the focus on mainland Southeast Asia, we feel that changes to classification are premature. In addition, because this phylogeny reflects only the history of the chloroplast in a group known for extensive hybridization (cf. Estep et al. 2014 and below), it is only a partial picture of the evolutionary history.

If supported by data on nuclear genes, *A. burmanicus* may need to be removed from *Andropogon*. *Andropogon distachyos* is the type species of the genus *Andropogon* (Hitchcock and Green 1947; Jarvis 1992), which suggests that *Andropogon* s. s. will be the clade that is sister to *Hyparrhenia*. Likewise, *Hemisorghum* can probably be returned to *Sorghum*, to which is it morphologically similar, if future analyses confirm the placement shown here. Our data reaffirm the decision to combine *Chrysopogon* and *Vetiveria* Bory because of morphological overlap between *C. zizanioides* (previously known as *Vetiveria zizanioides* (L.) Nash) and other species of *Chrysopogon* (Celarier 1959; Veldkamp 1999; Skendzic et al. 2007; Kellogg 2015).

*Andropogon fastigiatus* has been placed in its own genus, *Diectomis*, removed from *Andropogon* on the basis of its single inflorescence axis and the large glumes on its pedicellate spikelets. Our data place it in a separate clade from *Andropogon* s. s., suggesting it is indeed misplaced and may be more closely related to *Schizachyrium*, to which it is morphologically similar.

The type species of *Schizachyrium* is *S. brevifolium*, and thus the name *Schizachyrium* will apply to the clade with reduced pedicellate spikelets that is sister to *Andropogon chinensis*. If the current topology is supported, then *S. tenerum* and *S. imberbe* would need to be transferred to another genus. Alternatively, *A. chinensis* and *A. fastigiatus* could be transferred to *Schizachyrium* to preserve monophyly.

Like generic limits, subtribal limits within Andropogoneae are still in flux. The most recent classifications by Kellogg (2015) and Soreng et al. (2015) differ both in the number of recognized subtribes and their circumscription. The difference in part is because Kellogg (2015) avoids monogeneric subtribes for genera that are not strongly placed in the phylogeny, whereas Soreng et al. (2015) use available subtribal names when possible. The core Andropogoneae was placed in a very broad Andropogoninae by Kellogg (2015) but divided by Soreng et al. (2015) into Andropogoninae and Anthistiriinae, although the circumscription of those subtribes was different from those suggested by Clayton and Renvoize (1986). Figure 1 shows two major clades within the core, which were also found by Estep et al. (2014) in their study of nuclear genes. Thus we suggest following Soreng et al. (2015) in recognizing

both Andropogoninae (*Andropogon*, *Schizachyrium*, and *Hyparrhenia*) and Anthistiriinae (*Themeda*, *Heteropogon*, and *Cymbopogon*) as making up the core Andropogoneae.

Because of the sample presented here, our data do not test monophyly of subtribes Ischaeminae (including *Dimeria*, and *Ischaemum* L.) or Germainiinae (including *Germainia* and *Apocopsis* Nees). The monotypic genus *Kerriochloa* was placed in Ischaeminae by Clayton and Renvoize (1986) and by Soreng et al. (2015), who distinguished it from other genera based on the presence of a spatheole wrapped around the single inflorescence axis. However, Kellogg (2015) placed the genus *incertae sedis* in Andropogoneae, a placement we feel is still appropriate based on its lack of close relatives in the current tree.

We confirmed the traditional subtribe “Saccharinae” as polyphyletic (Hodkinson et al. 2002; Welker et al. 2016), with scattered positions of *Eulalia*, *Eulaliopsis*, and *Pogonatherum*. Prior to this study, Teerawatananon et al. (2011), GPWG II (2012), Estep et al. (2014), and Welker et al. (2015, 2016) also demonstrated that *Pogonatherum* is linked with subtribe Germainiinae (*Germainia* and *Apocopsis*). Furthermore, the sister relationship of subtribe Sorghinae, represented here by *Hemisorghum*, *Pseudosorghum*, and *Sorghum*, to the core Andropogoneae was also verified in this study (Soreng et al. 2015).

*Chrysopogon* and *Eriochrysis* are placed *incertae sedis* in both current classifications (Soreng et al. 2015; Kellogg 2015).

**Morphological Character Evolution**—The morphological characters traditionally used to distinguish *Andropogon* from *Schizachyrium*, as well as to characterize other genera, vary not only in those two genera but among many members of the tribe. The number of primary branches in the inflorescence (i.e. the products of the IM) and the number of orders of branching (i.e. the products of the branch meristems) are variable enough that ancestral state reconstructions are ambiguous throughout the tree, precluding any reliable estimates of the number of changes. Not only do these characters fail to distinguish species classified in *Andropogon* from those in *Schizachyrium* (see for example *Andropogon fastigiatus*), but they also are highly variable within other clades as well (Fig. 2).

The character state at the base of the tree is ambiguous for both characters. However, our optimization suggests a high probability that the primary branch number is zero at the deepest nodes of the phylogeny, whereas the number of orders of branching has a high probability of being one at the deepest nodes, implying that the number of primary branches must be more than zero. The apparent contradiction between the characters is almost certainly an artifact. The two characters have different numbers of states and are estimated to be evolving under different modes of evolution. Also, they are known to be genetically separable and to vary independently (Kellogg et al. 2013). While the common ancestor cannot have had both an unbranched inflorescence and a single order of branching, analyses of both characters are consistent in suggesting that highly branched inflorescences (“panicles”) are derived.

Likewise, the extent of development of the pedicellate spikelet and its size relative to the sessile spikelet are variable, implying that they are easily modified in evolutionary time. Pedicellate spikelets are rudimentary in a number of species of *Schizachyrium*, and are lost entirely in *Eremochloa*, *Coix*, and the female inflorescences of *Zea*. Only in the genus *Dimeria* is the sessile spikelet lost.

The sex expression of the two spikelets varies among genera. In some genera (e.g. *Miscanthus* Andersson, *Eriochrysis*), both the sessile and pedicellate spikelets bear bisexual florets. The lack of differentiation between sessile and pedicellate spikelets is the characteristic that Hartley (1958) interpreted to be primitive. More commonly (e.g. *Andropogon*, *Schizachyrium*), the sessile spikelet bears a bisexual floret whereas the pedicellate spikelet is staminate or sterile. In a handful of taxa (e.g. *Germainia*), the situation is reversed and the pedicellate spikelet is pistillate with the sessile one staminate or sterile (Clayton and Renvoize 1986).

Some genera (e.g. *Hyparrhenia*) also bear one or more pairs of staminate spikelets at the bases of the inflorescence branches; such pairs are known as homogamous pairs (Clayton 1990). Homogamous spikelet pairs are found in the clade of *Cymbopogon*, *Dichanthium*, *Heteropogon*, and *Themeda* (subtribe Anthistiriinae). Outside this lineage, the pairs are also found in *Hyparrhenia* and *Germainia*. Consistent with all its closely related species, *Parahyparrhenia siamensis* lacks homogamous pairs. However, the other five (unsampled) species of *Parahyparrhenia* A. Camus have them (Clayton 1969; Teerawatananon et al. 2013). Because we only sampled *P. siamensis*, our data may indicate that homogamous pairs arise independently in the *Parahyparrhenia-Eulalia contorta-Andropogon burmanicus* clade. Alternatively, *Parahyparrhenia* may be polyphyletic, and the unsampled species may all belong in the *Hyparrhenia* clade.

**Hybridization and Polyploidy as Explanations for the Phylogenetic Pattern**—Estep et al. (2014) documented extensive hybridization and polyploidy in Andropogoneae. Their phylogenetic data showed that a minimum of 1/3 of the species were allopolyploids and that most formed in independent polyploidization events. When combined with facultative apomixis, which occurs in some genera of Andropogoneae, the possibility exists of a highly reticulate evolutionary pattern, as previously demonstrated for *Bothriochloa*, *Capillipedium* and *Dichanthium* (Estep et al. 2014). Thus, many of the surprising placements of taxa we find here could be explained by hybridization, which can in turn lead to changes in distribution and ecological tolerance. The chloroplast genome can identify only one parental genome of an allopolyploid species (Mason-Gamer 2007, 2013; Kellogg 2016), and low-copy nuclear genes will need to be employed to test our results.

The limitations of plastome sequences for uncovering evolutionary relationships are especially clear in the case of *S. sanguineum*. This species has been reported to have a great range of allopolyploid levels based on cytology and analysis of low-copy loci (Peichoto et al. 2011; Estep et al. 2014). The two independent *S. sanguineum* clades could represent distinct allopolyploidy events. For example, Old World *S. exile* might have donated the plastome to *S. sanguineum* populations in Central Thailand. Alternatively, the presence of two clades could indicate wide introgression, but this seems unlikely given the distant geographic origins of the two clades. A final possibility is that of cryptic species. Although the major morphological characters place all specimens in *S. sanguineum*, it is possible that there are subtle characters not yet identified that distinguish the two clades. Apomixis has been reported in *S. sanguineum* (Carman and Hatch 1982), and this could easily lead to subtle morphological distinctions among populations that are partially or wholly reproductively isolated.

The tribe Andropogoneae is a primary component of grasslands and savannahs in the Americas and Africa, where members of the tribe tend to grow in areas with slightly higher precipitation than those of the drought-tolerant Chloridoideae and less tree cover than  $C_3$  tropical species in Paniceae (Gibson 2008; Bocksberger et al. 2016). However, Hartley (1958) recognized southeastern Asia as a center of diversity for Andropogoneae, characterized in part by the presence of multiple genera including *Sorghum*, *Miscanthus*, *Chrysopogon*, *Schizachyrium*, and *Andropogon*. Here, we have shown that cryptic diversity exists in mainland Southeast Asia, exemplified by *Andropogon burmanicus*, *Eulalia contorta* and *Parahyparrhenia siamensis*, by identifying a previously unidentified lineage in Andropogoneae with inflorescence characters that appear convergent with those of well-established genera. This study suggests that Southeast Asia may indeed harbor unique diversity in Andropogoneae and merits deeper study including characterization of polyploidy and hybridization through the use of low copy nuclear loci. Further implications of the phylogeny suggest that key characters for identification in Andropogoneae may be the result of convergent evolution and require revised taxonomic treatment.

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#### LITERATURE CITED

- Anderson, J. G. 1966. The genus *Andropogon* in Southern Africa. *Bothalia* 9: 5–30.
- Arthan, W., M. R. McKain, P. Traiperm, C. A. D. Welker, J. K. Teisher, and E. A. Kellogg. 2017. Data from: Phylogenomics of Andropogoneae (Panicoideae: Poaceae) of mainland Southeast Asia. Dryad Digital Repository. <http://dx.doi.org/10.5061/dryad.p01f4>.
- Barker, D., A. Meade, and M. Pagel. 2007. Constrained models of evolution lead to improved prediction of functional linkage from correlated gain and loss of genes. *Bioinformatics* 23: 14–20.
- Barkworth, M. E. 2003. Andropogoneae Dumort. Pp. 602–704 in *Flora of North America North of Mexico*, vol. 25. Magnoliophyta: Commelinidae (in part): Poaceae, part 2, eds. M. E. Barkworth, K. M. Capels, S. Long, and M. B. Piep. New York: Oxford University Press.
- Barrett, C. F., W. J. Baker, J. R. Corner, J. G. Conran, S. C. Lahmeyer, J. H. Leebens-Mack, J. Li, G. S. Lim, D. R. Mayfield-Jones, L. Perez, J. Medina, J. C. Pires, C. Santos, D. W. Stevenson, W. B. Zomlefer, and J. I. Davis. 2016. Plastid genomes reveal support for deep phylogenetic relationships and extensive rate variation among palms and other commelinid monocots. *The New Phytologist* 209: 855–870.
- Bocksberger, G., J. Schnitzler, C. Chatelain, P. Daget, T. Janssen, M. Schmidt, A. Thiombiano, and G. Zizka. 2016. Climate and the distribution of grasses in West Africa. *Journal of Vegetation Science* 27: 306–317.
- Bolger, A. M., M. Lohse, and B. Usadel. 2014. Trimmomatic: A flexible trimmer for Illumina sequence data. *Bioinformatics* 30: 2114–2120.
- Bor, N. L. 1960. *The grasses of Burma, Ceylon, India, and Pakistan (excluding Bambuseae)*. New York: Pergamon Press.
- Bouchenak-Khelladi, Y., N. Salamin, V. Savolainen, F. Forest, M. van der Bank, M. W. Chase, and T. R. Hodkinson. 2008. Large multi-gene phylogenetic trees of the grasses (Poaceae): Progress towards complete tribal and generic level sampling. *Molecular Phylogenetics and Evolution* 47: 488–505.
- Buitenhuis, A. G. and J. F. Veldkamp. 2001. Revision of *Eremochloa* (Gramineae-Andropogoneae-Rottboelliinae). *Blumea* 46: 399–420.
- Burke, S. V., W. P. Wysocki, F. O. Zuloaga, J. M. Craine, J. C. Pires, P. P. Edger, D. Mayfield-Jones, L. G. Clark, S. A. Kelchner, and M. R. Duvall. 2016. Evolutionary relationships in panicoid grasses based on plastome phylogenomics (Panicoideae: Poaceae). *BMC Plant Biology* 16: 140.

- Campbell, C. S. and P. G. Windisch. 1986. Chromosome numbers and their taxonomic implications for eight Brazilian Andropogons (Poaceae: Andropogoneae). *Brittonia* 38: 411–414.
- Camus, E. G. and A. Camus. 1922. *Flore générale de l'Indo-Chine* 1(3). Masson, Paris, France.
- Carbonell-Caballero, J., R. Alonso, V. Ibañez, J. Terol, M. Talon, and J. Dopazo. 2015. A phylogenetic analysis of 34 chloroplast genomes elucidated the relationships between wild and domestic species within the genus *Citrus*. *Molecular Biology and Evolution* 32: 2015–2035.
- Carman, J. G. and S. L. Hatch. 1982. Aposporous apomixis in *Schizachyrium* (Poaceae: Andropogoneae). *Crop Science* 22: 1252–1255.
- Celarié, R. P. 1959. Cytotaxonomy of the Andropogoneae. IV. Subtribe Sorgheae. *Cytologia* 24: 285–303.
- Chen, S., S. Bixing, S. M. Phillips, and S. A. Renvoize. 2006. Tribe Andropogoneae. Pp. 570–573 in *Flora of China*, vol. 22, Poaceae, eds. W. Zhengyi, P. H. Raven, and H. Deyuan. Beijing and St. Louis, Missouri: Science Press and Missouri Botanical Garden.
- Chen, C. H., J. F. Veldkamp, and C. S. Kuoh. 2012. Taxonomic revision of *Microstegium* s. str. (Andropogoneae, Poaceae). *Blumea* 57: 160–189.
- Christin, P. A., E. Samaritani, B. Petitpierre, N. Salamin, and G. Besnard. 2009. Evolutionary insights on C<sub>4</sub> photosynthetic subtypes in grasses from genomics and phylogenetics. *Genome Biology and Evolution* 1: 221–230.
- Clayton, W. D. 1964. Studies in the Gramineae: V. *Kew Bulletin* 17: 465–470.
- Clayton, W. D. 1969. *A revision of the genus Hyparrhenia*. Surrey, U. K.: Royal Botanic Gardens, Kew.
- Clayton, W. D. 1972. The awned genera of Andropogoneae. Studies in the Gramineae: XXXI. *Kew Bulletin* 27: 457–474.
- Clayton, W. D. 1990. The spikelet. Pp. 32–49 in *Reproductive versatility in the grasses*, ed. G. P. Chapman. Cambridge, U. K.: Cambridge University Press.
- Clayton, W. D. and S. A. Renvoize. 1986. *Genera graminum*. London: Her Majesty's Stationery Office.
- Clayton, W. D., M. S. Vorontsova, K. T. Harman, and H. Williamson. 2006. onwards. GrassBase – The online world grass flora. <http://www.kew.org/data/grasses-db.html>.
- Cotton, J. L., W. P. Wysocki, L. G. Clark, S. A. Kelchner, J. C. Pires, P. P. Edger, D. Mayfield-Jones, and M. R. Duvall. 2015. Resolving deep relationships of PACMAD grasses: A phylogenomic approach. *BMC Plant Biology* 15: 178.
- Doebley, J. 1990. Molecular evidence and the evolution of maize. *Economic Botany* 44: 6–27.
- Doyle, J. J. and J. L. Doyle. 1987. A rapid DNA isolation procedure for small quantities of fresh leaf tissue. *Phytochemical Bulletin* 19: 11–15.
- Doyle, J. J. and J. L. Doyle. 1990. Isolation of plant DNA from fresh tissue. *Focus (San Francisco, Calif.)* 12: 13–15.
- Doust, A. N., K. M. Devos, M. Gadberry, M. D. Gale, and E. A. Kellogg. 2005. The genetic basis for inflorescence variation between foxtail and green millet (Poaceae). *Genetics* 169: 1659–1672.
- Estep, M. C., M. R. McKain, D. Vela Diaz, J. Zhong, J. G. Hodge, T. R. Hodgkinson, D. J. Layton, S. T. Malcomber, R. Pasquet, and E. A. Kellogg. 2014. Allopolyploidy, diversification, and the Miocene grassland expansion. *Proceedings of the National Academy of Sciences USA* 111: 15149–15154.
- Estes, J. R. and R. J. Tylr. 1976. *The grasses and grasslands of Oklahoma: Status of current research on the native and introduced grasses of the state: A symposium*. Ardmore, Oklahoma: Samuel Roberts Noble Foundation.
- Gibson, D. J. 2008. *Grasses and grassland ecology*. Oxford, U. K.: Oxford University Press.
- Gilliland, H. B. 1971. *A revised flora of Malaya: an illustrated systematic account of the Malayan flora, including commonly cultivated plants*. Volume III, Grasses of Malaya. Singapore: The government printing office.
- Giussani, L. M., J. H. Cota-Sánchez, F. O. Zuloaga, and E. A. Kellogg. 2001. A molecular phylogeny of the grass subfamily Panicoideae (Poaceae) shows multiple origins of C<sub>4</sub> photosynthesis. *American Journal of Botany* 88: 1993–2012.
- Givnish, T. J., D. Spalink, M. Ames, S. P. Lyon, S. J. Hunter, A. Zuluaga, W. J. D. Iles, M. A. Clements, M. T. K. Arroya, J. Leebens-Mack, L. Endara, R. Kriebel, K. M. Neubig, W. M. Whitten, N. H. Williams, and K. M. Cameron. 2015. Orchid phylogenomics and multiple drivers of their extraordinary diversification. *Proceedings of the Royal Society B: Biological Sciences* 282: 20151553.
- Gould, F. W. 1967. The genus *Andropogon* in the United States. *Brittonia* 19: 70–76.
- GPWG (Grass Phylogeny Working Group). 2001. Phylogeny and subfamilial classification of the grasses (Poaceae). *Annals of the Missouri Botanical Garden* 88: 373–457.
- GPWG II (Grass Phylogeny Working Group II). 2012. New grass phylogeny resolves deep evolutionary relationships and discovers C<sub>4</sub> origins. *The New Phytologist* 193: 304–312.
- Hand, M. L., G. C. Spangenberg, J. W. Forster, and N. O. I. Cogan. 2013. Plastome sequence determination and comparative analysis for members of the *Lolium-Festuca* grass species complex. *G3-Genes Genomes Genetics* 3: 607–616.
- Hartley, W. 1950. The global distribution of tribes of the Gramineae in relation to historical and environmental factors. *Australian Journal of Agricultural Research* 1: 355–372.
- Hartley, W. 1958. Studies on the origin, evolution and distribution of the Gramineae. I. The tribe Andropogoneae. *Australian Journal of Botany* 6: 116–128.
- Hitchcock, A. S. and M. L. Green. 1947. Supplement: Species Lectotypicae Generum Linnaei. *Brittonia* 6: 114–118.
- Hodkinson, T. R., M. W. Chase, M. D. Lledó, N. Salamin, and S. A. Renvoize. 2002. Phylogenetics of *Miscanthus*, *Saccharum* and related genera (Saccharinae, Andropogoneae, Poaceae) based on DNA sequences from ITS nuclear ribosomal DNA and plastid *trnL* intron and *trnL-F* intergenic spacers. *Journal of Plant Research* 115: 381–392.
- Jannink, T. A. and J. F. Veldkamp. 2002. Revision of Chionachninae (Gramineae: Andropogoneae). *Blumea* 47: 545–580.
- Jarvis, C. E. 1992. Seventy-two proposals for the conservation of types of selected Linnaean generic names, the report of Subcommittee 3C on the lectotypification of Linnaean generic names. *Taxon* 41: 552–583.
- Katoh, K., K. I. Kuma, H. Toh, and T. Miyata. 2005. MAFFT version 5: Improvement in accuracy of multiple sequence alignment. *Nucleic Acids Research* 33: 511–518.
- Kearse, M., R. Moir, A. Wilson, S. Stones-Havas, M. Cheung, S. Sturrock, S. Buxton, A. Cooper, S. Markowitz, C. Duran, T. Thierer, B. Ashton, P. Meintjes, and A. Drummond. 2012. Geneious basic: An integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics* 28: 1647–1649.
- Kellogg, E. A. 2000. Molecular and morphological evolution in the Andropogoneae. Pp. 149–158 in *Grasses: Systematics and evolution*, eds. S. W. L. Jacobs. and J. Everett. Melbourne, Australia: CSIRO.
- Kellogg, E. A. 2015. Tribe Andropogoneae Dumort. Pp. 289–314 in *The families and genera of vascular plants* vol. XIII, ed. K. Kubitski. Cham, Switzerland: Springer International.
- Kellogg, E. A. 2016. Has the connection between polyploidy and diversification actually been tested?. *Current Opinion in Plant Biology* 30: 25–32.
- Kellogg, E. A., P. E. A. S. Camara, P. J. Rudall, P. Ladd, S. T. Malcomber, C. J. Whipple, and A. N. Doust. 2013. Early inflorescence development in the grasses (Poaceae). *Frontiers in Plant Science* 4: 250.
- Langmead, B. and S. Salzberg. 2012. Fast gapped-read alignment with Bowtie 2. *Nature Methods* 9: 357–359.
- Lazarides, M. 1980. *Phanerogamarum monographiae. Tomus XII: The tropical grasses of Southeast Asia (excluding bamboos)*. Germany: Strauss and Cramer.
- Lewis, P. O. 2001. A likelihood approach to estimating phylogeny from discrete morphological character data. *Systematic Biology* 50: 913–925.
- Marçais, G. and C. Kingsford. 2011. A fast, lock-free approach for efficient parallel counting of occurrences of *k*-mers. *Bioinformatics* 27: 764–770.
- Mason-Gamer, R. J. 2007. Allopolyploids of the genus *Elymus* (Triticeae, Poaceae): A phylogenetic perspective. *Aliso* 23: 372–379.
- Mason-Gamer, R. J. 2013. Phylogeny of a genomically diverse group of *Elymus* (Poaceae) allopolyploids reveals multiple levels of reticulation. *PLoS One* 8: e78449.
- Mason-Gamer, R. J., C. F. Weil, and E. A. Kellogg. 1998. Granule-bound starch synthase: structure, function, and phylogenetic utility. *Molecular Biology and Evolution* 15: 1658–1673.
- Mathews, S., R. E. Spangler, R. J. Mason-Gamer, and E. A. Kellogg. 2002. Phylogeny of Andropogoneae inferred from phytochrome B, GBSI, and *ndhF*. *International Journal of Plant Sciences* 163: 441–450.
- McKain, M. R., R. H. Hartssock, M. M. Wohl, and E. A. Kellogg. 2016a. Verdant: Automated annotation, alignment and phylogenetic analysis of whole chloroplast genomes. *Bioinformatics* 33: 130–132.
- McKain, M. R., J. R. McNeal, P. R. Kellar, L. E. Eguarte, J. C. Pires, and J. Leebens-Mack. 2016b. Timing of rapid diversification and convergent origins of active pollination within Agavoideae (Asparagaceae). *American Journal of Botany* 103: 1717–1729.
- Miller, M. A., W. Pfeiffer, and T. Schwartz. 2010. Creating the CIPRES science gateway for inference of large phylogenetic trees. *Proceedings*



- of the Gateway Computing Environments Workshop (GCE) in New Orleans, Louisiana.
- Nagahama, N. and G. A. Norrmann. 2012. Review of the genus *Andropogon* (Poaceae: Andropogoneae) in America based on cytogenetic studies. *Le Journal de Botanique* 2012: 1–9.
- Nanakorn, W. 1990. *The floristic treatment of the tribe Andropogoneae: subtribes Andropogoninae, Anthistirinae and Saccharinae (Gramineae) for the flora of Thailand*. Ph.D. dissertation. New York: The City University of New York.
- Neamsuvan, O., J. F. Veldkamp, and T. Seelanan. 2009. A revision of *Hemisorghum* (Poaceae, Sorghineae) in Thailand. *Gardens' Bulletin Singapore* 61: 145–149.
- Nurk, S., A. Bankevich, D. Antipov, A. A. Gurevich, A. Korobeynikov, A. Lapidus, A. D. Priblensky, A. Pyshkin, A. Sirotkin, Y. Sirotkin, R. Stepanauskas, S. R. Clingenpeel, T. Woyke, J. S. McLean, R. Laskin, G. Tesler, M. A. Alekseyev, and P. A. Pevzner. 2013. Assembling genomes and mini-metagenomes from highly chimeric reads. *Research in Computational Molecular Biology* 7821: 158–170.
- Palmer, J. D. 1985. Comparative organization of chloroplast genomes. *Annual Review of Genetics* 19: 325–354.
- Paradis, E., J. Claude, and K. Strimmer. 2004. APE: Analyses of phylogenetics and evolution in R language. *Bioinformatics* 20: 289–290.
- Peichoto, M. C. 2010. Revisión taxonómica de las especies del género *Schizachyrium* (Poaceae: Andropogoneae) de Sudamérica. *Candollea (Genève)* 65: 301–345.
- Peichoto, M. C., F. Galdeano, M. M. Arbo, and V. G. S. Neffa. 2011. Chromosome number and ploidy level of some South American species of *Schizachyrium* (Poaceae, Andropogoneae). *Plant Systematics and Evolution* 292: 63–71.
- Rambaut, A. 2014. FigTree, version 1.4.2 for Windows. Computer program and documentation distributed by the author. <http://tree.bio.ed.ac.uk/software/figtree/> (last accessed 21 March 2016).
- Revell, L. J. 2012. phytools: An R package for phylogenetic comparative biology (and other things). *Methods in Ecology and Evolution* 3: 217–223.
- Ronquist, F., M. Teslenko, P. van der Mark, D. L. Ayres, A. Darling, S. Höhna, B. Larget, L. Liu, M. A. Suchard, and J. P. Huelsenbeck. 2012. MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology* 61: 539–542.
- Ruhfel, B. R., M. A. Gitzendanner, P. S. Soltis, D. E. Soltis, and J. G. Burleigh. 2014. From algae to angiosperms – inferring the phylogeny of green plants (Viridiplantae) from 360 plastid genomes. *BMC Evolutionary Biology* 14: 23.
- Shouliang, C. and S. M. Phillips. 2006. *Andropogon*. Pp. 570–573 in *Flora of China* vol. 22, eds. W. Zhengyi, P. H. Raven, and H. Deyuan. Beijing and St. Louis, Missouri: Science Press and Missouri Botanical Garden.
- Simon, B. K. 2007. Grass phylogeny and classification: Conflicts of morphology and molecules. *Aliso* 23: 259–266.
- Skendzic, E. M., J. T. Columbus, and R. Ceros-Tlatilpa. 2007. Phylogenetics of Andropogoneae (Poaceae: Panicoideae) based on nuclear ribosomal internal transcribed spacer and chloroplast *trnL-F* sequences. *Aliso* 23: 530–544.
- Soreng, R. J., P. M. Peterson, K. Romaschenko, G. Davidse, F. O. Zuloaga, E. J. Judziewicz, T. S. Figueiras, J. I. Davis, and O. Morrone. 2015. A worldwide phylogenetic classification of the Poaceae. *Journal of Systematics and Evolution* 53: 117–137.
- Spangler, R., B. Zaitchik, E. Russo, and E. A. Kellogg. 1999. Andropogoneae evolution and generic limits in *Sorghum* (Poaceae) using *ndhF* sequences. *Systematic Botany* 24: 267–281.
- Stamatakis, A. 2006. RAxML-VI-HPC: Maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics* 22: 2688–2690.
- Steele, P. R., K. L. Hertweck, D. Mayfield-Jones, M. R. McKain, J. Leebens-Mack, and J. C. Pires. 2012. Quality and quantity of data recovered from massively parallel sequencing: Examples in Asparagales and Poaceae. *American Journal of Botany* 99: 330–348.
- Stull, G. W., M. J. Moore, V. S. Mandala, N. A. Douglas, H.-R. Kates, X. Qi, S. F. Brockington, P. S. Soltis, D. E. Soltis, and M. A. Gitzendanner. 2013. A targeted enrichment strategy for massively parallel sequencing of Angiosperm plastid genomes. *Applications in Plant Sciences* 1: 1200497.
- Teerawatananon, A., V. Boontia, B. Chantarasuwan, T. R. Hodkinson, and S. Sungkaew. 2014. A taxonomic revision of the genus *Dimeria* (Poaceae: Panicoideae) in Thailand. *Phytotaxa* 186: 137–147.
- Teerawatananon, A., S. W. L. Jacobs, and T. R. Hodkinson. 2011. Phylogenetics of Panicoideae (Poaceae) based on chloroplast and nuclear DNA sequences. *Telopea* 13: 115–142.
- Teerawatananon, A., S. Sungkaew, V. Boontia, and T. R. Hodkinson. 2013. A taxonomic revision of *Germainia* (Andropogoneae: Poaceae) in Thailand. *Blumea* 57: 221–228.
- Thompson, B. 2014. Genetic and hormonal regulation of maize inflorescence development. *Advances in Botanical Research* 72: 263–296.
- Traiperm, P. 2007. *Systematics of Ischaeminae and Rottboelliinae (Poaceae) in Thailand*. Ph.D. dissertation. Bangkok: Chulalongkorn University.
- Türpe, A. M. 1984. Revision of the South American species of *Schizachyrium* (Gramineae). *Kew Bulletin* 39: 169–178.
- Ueno, O. 1995. Occurrence of distinctive cells in leaves of C<sub>4</sub> species in *Arthraxon* and *Microstegium* (Andropogoneae-Poaceae) and the structural and immunocytochemical characterization of these cells. *International Journal of Plant Sciences* 156: 270–289.
- Vallejo-Marín, M., A. M. Cooley, M. Y. Lee, M. Folmer, M. R. McKain, and J. R. Puzey. 2016. Strongly asymmetric hybridization barriers shape the origin of a new polyploid species and its hybrid ancestor. *American Journal of Botany* 103: 1272–1288.
- Van Welzen, P. C. 1981. A taxonomic revision of the genus *Arthraxon* Beauv. (Gramineae). *Blumea* 27: 255–300.
- Veldkamp, J. F. 1999. A revision of *Chrysopogon* Trin. including *Vetiveria* Bory (Poaceae) in Thailand and Malesia with notes on some other species from Africa and Australia. *Austrobaileya* 5: 503–533.
- Veldkamp, J. F. 2003. Miscellaneous notes on Thai Gramineae. *Blumea* 48: 495–501.
- Veldkamp, J. F., M. E. B. van den Boogaart, J. Heidweiller, M. A. F. van der Klaauw, R. de Koning, A. R. Kraaijeveld, M. S. M. Sosef, and R. C. W. Strucker. 2013. A revision of *Mnesithea* (Gramineae – Rottboelliinae) in Malesia and Thailand. *Blumea* 58: 277–292.
- Veldkamp, J. F., A. Teerawatananon, and S. Sungkaew. 2015. A revision of *Garnotia* (Gramineae) in Malesia and Thailand. *Blumea* 59: 229–237.
- Vorontsova, M. S., G. Ratovonirina, and T. Randriamboavonjy. 2013. Revision of *Andropogon* and *Diectomis* (Poaceae: Sacchareae) in Madagascar and the new *Andropogon itremoensis* from the Itremo Massif. *Kew Bulletin* 68: 1–15.
- Washburn, J. D., J. C. Schnable, G. Davidse, and J. C. Pires. 2015. Phylogeny and photosynthesis of the grass tribe Paniceae. *American Journal of Botany* 102: 1493–1505.
- Watson, L. and M. J. Dallwitz. 1992. *The grass genera of the world*. Wallingford, Oxon, U. K.: CAB International.
- Welker, C. A. D. and H. M. Longhi-Wagner. 2012. The genera *Eriochrysis* P. Beauv., *Imperata* Cirillo and *Saccharum* L. (Poaceae – Andropogoneae – Saccharinae) in the state of Rio Grande do Sul, Brazil. *Brazilian Journal of Botany* 35: 87–105.
- Welker, C. A. D., T. T. Souza-Chies, H. M. Longhi-Wagner, M. C. Peichoto, M. R. McKain, and E. A. Kellogg. 2015. Phylogenetic analysis of *Saccharum* s.l. (Poaceae; Andropogoneae), with emphasis on the circumscription of the South American species. *American Journal of Botany* 102: 248–263.
- Welker, C. A. D., T. T. Souza-Chies, H. M. Longhi-Wagner, M. C. Peichoto, M. R. McKain, and E. A. Kellogg. 2016. Multilocus phylogeny and phylogenomics of *Eriochrysis* P. Beauv. (Poaceae-Andropogoneae): Taxonomic implications and evidence of interspecific hybridization. *Molecular Phylogenetics and Evolution* 99: 155–167.
- Zanin, A. and H. M. Longhi-Wagner. 2011. Revisão de *Andropogon* (Poaceae – Andropogoneae) para o Brasil. *Rodriguésia* 62: 171–202.
- Zuloaga, F. O., O. Morrone, G. Davidse, and S. J. Pennington. 2007. Classification and biogeography of Panicoideae (Poaceae) in the New World. *Aliso* 23: 503–529.

APPENDIX 1. Taxa included in this study, collection localities, voucher specimens, herbarium location, and GenBank accession number.

*Andropogon abyssinicus* R. Br. ex Fresen, Nairobi, Kenya, *Pasquet1097* (MO), KY596148. *Andropogon burmanicus* Bor, Ubon Ratchathani, Thailand, *Arthan 071* (BKF), KY596164, *Traiperm 572* (BKF, MO), KY596167. *Andropogon chinensis* (Nees) Merr., Chiang Mai, Thailand, *Arthan 042* (BKF), KY596126, *Chaiyaphum*, Thailand, *Arthan 062* (BKF), KY596121, *Arthan 063* (BKF), KY596133. *Andropogon distachyos* L., Chiang Mai, Thailand, *Arthan 050* (BKF), KY596170. *Andropogon fastigiatus* Sw., Chaiyaphum, Thailand, *Arthan 009* (BKF), KY596152, Chiang Mai, Thailand, *Arthan 024* (BKF), KY596180, Loei, Thailand, *Arthan 031* (BKF), KY596118, Phetchabun, Thailand, *Arthan 043* (BKF), KY596144. *Arthraxon hispidus* (Thunb.) Makino, Chiang Mai, Thailand, *Arthan 023*, (BKF), KY596179. *Arthraxon lanceolatus* (Roxb.) Hochst., Loei, Thailand, *Arthan 001* (BKF), KY596127, Phetchabun, Thailand, *Arthan 059* (BKF), KY596185. *Arthraxon microphyllus* (Trin.) Hochst., Chiang Mai, Thailand, *Traiperm 537* (BKF), KY596183.

*Arthraxon prionodes* (Steud.) Dandy, Xizang, China, Kellogg PI 659331 (MO), KY596138. *Chrysopogon gryllus* (L.) Trin., Skopje, Republic of Macedonia, Kellogg PI 250984 (A), KY596161. *Chrysopogon orientalis* (Desv.) A. Camus, Ubon Ratchathani, Thailand, *Traiperm* 578 (BKF), KY596177. *Chrysopogon serrulatus* Trin., Bannu, Pakistan, Kellogg PI 219580 (A, MO), NC\_029884. *Chrysopogon zizanioides* (L.) Roberty, Cultivated specimen from Maryland, USA, Kellogg Vet-MRL-001 (MO), KY596158. *Coix lacryma-jobi* L., Chaiyapum, Thailand, *Arthan* 072 (BKF), KY596160. *Cymbopogon flexuosus* (Nees ex Steud.) Will. Watson, Chaiyapum, Thailand, *Arthan* 027 (BKF), KY596169. *Dichanthium sericeum* (R. Br.) A. Camus, Nong Kai, Thailand, *Traiperm* 571 (BKF), KY596128. *Dimeria ornithopoda* Trin., Ubon Ratchathani, Thailand, *Traiperm* 575 (BKF), KY596130. *Eremochloa ciliaris* (L.) Merr., Chaiyapum, Thailand, *Traiperm* 524 (BKF), KY596146. *Eremochloa eriopoda* C.E. Hubb., Udon Thani, Thailand, *Traiperm* 591 (BKF), KY596134. *Eriochrysis cayennensis* P. Beauv., São Francisco de Paula, Rio Grande do Sul, Brazil, *Welker* 365 (ICN), NC\_029882, Palmeira, Paraná, Brazil, *Welker* 519 (ICN), KU961861. *Eriochrysis laxa* Swallen, São Borja, Rio Grande do Sul, Brazil, *Welker* 489 (ICN), NC\_029883. *Eriochrysis villosa* Swallen, São Luiz Gonzaga, Rio Grande do Sul, Brazil, *Welker* 481B (ICN), KU961860. *Eulalia contorta* (Brongn.) Kuntze, Ubon Ratchathani, Thailand, *Traiperm* 573 (BKF), KY596143. *Eulalia siamensis* Bor, Chiang Mai, Thailand, *Traiperm* 557 (BKF), KY596149. *Eulaliopsis binata* (Retz.) C.E. Hubb., Chiang Mai, Thailand, *Traiperm* 567 (BKF), KY596182. *Garnotia tenella* (Arn. ex Miq.) Janowski, Chiang Mai, Thailand, *Traiperm* 552 (BKF), KY596184. *Garnotia thailandica* Gould, Chiang Mai, Thailand, *Traiperm* 535 (BKF), KY596171. *Germainia capitata* Balansa & Poitr., Chaiyapum, Thailand, *Arthan* 028 (BKF), KY596175. *Hemisorghum mekongense* (A. Camus) C.E. Hubb., Nong Kai, Thailand, *Traiperm* 569 (BKF), KY596132. *Heteropogon contortus* (L.) P. Beauv. ex Roem. & Schult., Chiang Mai, Thailand, *Arthan* 035 (BKF), KY596145. *Heteropogon triticeus* (R. Br.) Stapf ex Craib, Chiang Mai, Thailand, *Arthan* 034 (BKF), KY596142, *Traiperm* 534 (BKF), KY596159, Phetchabun, Thailand, *Arthan* 017 (BKF), KY596153, *Arthan* 046 (BKF), KY596176. *Hyparrhenia diplandra* (Hack.) Stapf, Phetchabun, Thailand, *Arthan* 012 (BKF), KY596163. *Hyparrhenia rufa* (Nees) Stapf, Chiang Mai, Thailand, *Arthan* 045 (BKF), KY596156, Loei, Thailand, *Arthan* 068 (BKF), KY596119. *Kerriochloa siamensis* C.E. Hubb., Ubon Ratchathani, Thailand, *Traiperm* 580 (BKF), KY596120, *Traiperm* 581 (BKF), KY596117. *Mnesithea helferi* (Hook. f.) de Koning & Sosef, Ubon Ratchathani, Thailand, *Traiperm* 574 (BKF), KY596162. *Parahyparrhenia siamensis* Clayton, Ubon Ratchathani, Thailand, *Traiperm* 583 (BKF), KY596155. *Pogonatherum panicum* (Lam.) Hack., Cultivated; Univ. of Missouri, St. Louis, Clark s.n. (MO), NC\_029881. *Polytoca digitata* (L. f.) Druce, Phetchabun, Thailand, *Arthan* 054 (BKF), KY596178, *Arthan* 060 (BKF), KY596173. *Pseudosorghum fasciculare* (Roxb.) A. Camus, Loei, Thailand, *Arthan* 067 (BKF), KY596157, Phetchabun, Thailand, *Traiperm* 512 (BKF), KY596139. *Schizachyrium brevifolium* (Sw.) Nees ex Buse, Chiang Mai, Thailand, *Arthan* 010 (BKF), KY596122, Loei, Thailand, *Arthan* 013 (BKF), KY596168, Phetchabun, Thailand, *Arthan* 019 (BKF), KY596150, *Arthan* 030 (BKF), KY596151, Chaiyapum, Thailand, *Arthan* 041 (BKF), KY596135. *Schizachyrium exile* (Hochst.) Pilg., Chiang Mai, Thailand, *Arthan* 047 (BKF), KY596147. *Schizachyrium imberbe* A. Camus, Mercedes, Corrientes, Argentina, *Welker & Peichoto* 564 (ICN), KY596174. *Schizachyrium sanguineum* (Retz.) Alston, Loei, Thailand, *Arthan* 005 (BKF), KY596137, Phetchabun, Thailand, *Arthan* 011 (BKF), KY596140, *Arthan* 014 (BKF), KY596181, Loei, Thailand, *Arthan* 022 (BKF), KY596124, Chaiyapum, Thailand, *Arthan* 026 (BKF), KY596165, *Arthan* 033 (BKF), KY596141. *Schizachyrium scoparium* (Michx.) Nash, St. Louis, Missouri, United States, Kellogg V46 (MO), KY596154. *Schizachyrium spicatum* (Spreng.) Herter, Velázquez, Rocha, Uruguay, *Welker* 627 (ICN), KY596166. *Schizachyrium tenerum* Nees, Ituzaingó, Corrientes, Argentina, *Welker & Peichoto* 604 (ICN), KY596172. *Sorghum bicolor* (L.) Moench, Unknown, Cultivar BTx623, (Unknown), EF115542 (whole genome). *Themeda arguens* (L.) Hack., Loei, Thailand, *Arthan* 020 (BKF), KY596129. *Themeda arundinacea* (Roxb.) A. Camus, Chiang Mai, Thailand, *Arthan* 064 (BKF), KY596123. *Themeda triandra* Forssk., Sakon Nakorn, Thailand, *Arthan* 070 (BKF), KY596125. *Themeda villosa* (Poir.) A. Camus, Chaiyapum, Thailand, *Arthan* 065, (BKF), KY596131. *Zea mays* L., Unknown, Cultivar B73, (Unknown) X86563 (whole genome).

APPENDIX 2. Geographical distribution of taxa in this study, showing location in Southeast Asia. SC = Southern China; My = Myanmar; T = Thailand; L = Laos; V = Vietnam; Ca = Cambodia; PM = Peninsular Malaysia; I = Indonesia; Ph = Philippines. Data from Bor (1960), Camus and Camus (1922), Gililand (1971), Lazarides (1980), Nanakorn (1990), and from the following electronic resources: An online resource for monocot plants. (n.d.). Retrieved January 16, 2017, <http://e-monocot.org/>; Kew Herbarium Catalogue. (n.d.). Retrieved January 16, 2017,

from <http://apps.kew.org/herbcat/navigator.do>; Flora of China @ efloras.org. (n.d.). Retrieved January 16, 2017, [http://www.efloras.org/flora\\_page.aspx?flora\\_id=2&](http://www.efloras.org/flora_page.aspx?flora_id=2&); Tropicos - Home. (n.d.). Retrieved January 16, 2017, from <http://www.tropicos.org/>

**Andropogon L.:** *A. abyssinicus*, native to Africa; *A. burmanicus*, My, T, L, V; *A. chinensis*, SC, My, T, L, V, Ca; *A. distachyos*, T (introduced); *A. fastigiatus*, My, T, L, V, I, Ph. **Arthraxon P. Beauv.:** *A. hispidus*, SC, My, T, L, V, Ca, PM, I, Ph; *A. lanceolatus*, SC, My, T, V, PM, I; *A. microphyllus*, SC, T; *A. prionodes*, SC, My, T, V, PM. **Chrysopogon Trin.:** *C. gryllus*, SC, T; *C. orientalis*, SC, My, T, L, V, Ca, PM, I; *C. serrulatus*, My, T, PM, I; *C. zizanioides*, SC (introduced), My, T, L, V, Ca, PM (introduced), I, Ph. **Coix L.:** *C. lacryma-jobi*, SC, My, T, L, V, Ca, PM, I, Ph (introduced). **Cymbopogon Spreng.:** *C. flexuosus*, SC (introduced), My, T, V, PM (introduced), I (introduced). **Dichanthium Willemet:** *D. sericeum*, T, L, V, Ca, Ph. **Dimeria R. Br.:** *D. ornithopoda*, SC, My, T, L, V, Ca, PM, I, Ph. **Eremochloa Buse:** *E. ciliaris*, SC, My, T, L, V, Ca, PM, I, Ph; *E. eriopoda*, T, L, V, Ca, I. **Eriochrysis P. Beauv.:** *E. cayennensis* (native in America); *E. laxa* (native in America); *E. villosa* (native in America). **Eulalia Kunth:** *E. contorta*, SC, My, T, L, V, PM, I, Ph; *E. siamensis*, SC, My, T, L. **Eulaliopsis Honda:** *E. binata*, SC, My, T, L, Ph. **Garnotia Brongn.** (outgroup). **Germainia Balansa & Poitr.:** *G. capitata*, SC, T, V, I. **Heteropogon Pers.:** *H. contortus*, SC, My, T, V, PM, I, Ph; *H. triticeus*, My, T, L, V, PM, I, Ph. **Hemisorghum C.E. Hubb. ex Bor:** *H. mekongense*, My, T, L, V. **Hyparrhenia Andersson ex E. Fourn.:** *H. diplandra*, SC, T, V, I; *H. rufa*, SC, My, T, I (introduced), Ph (introduced). **Kerriochloa C.E. Hubb.:** *K. siamensis*, T, V, Ca. **Mnesithea Kunth:** *M. helferi*, My, T, V, Ca, PM. **Parahyparrhenia A. Camus:** *P. siamensis*, T, L. **Pogonatherum P. Beauv.:** *P. panicum*, SC, My, T, L, V, PM, I. **Polytoca R. Br.:** *P. digitata*, SC, My, T, V, Ca, PM, I, Ph. **Pseudosorghum A. Camus:** *P. fasciculare*, SC, My, T, V, Ph. **Schizachyrium Nees:** *S. brevifolium*, SC, My, T, L, V, PM, I, Ph; *S. exile*, My, T; *S. imberbe*, native to America; *S. sanguineum*, SC, My, T, L, V, PM, I, Ph; *S. scoparium*, native to America; *S. spicatum*, native to America; *S. tenerum*, native to America. **Sorghum Moench:** *S. bicolor*, native to Africa. **Themeda Forssk.:** *T. arguens*, My, T, L, V, PM, I; *T. arundinacea*, SC, My, T, L, V, Ca, PM, I, Ph; *T. triandra*, SC, My, T, L, V, PM, I, Ph; *T. villosa*, SC, My, T, L, V, Ca, PM, I, Ph. **Zea L.:** *Z. mays*, native to America.

APPENDIX 3. Characters and character states for morphological analysis. Characters are primary inflorescence branches, orders of inflorescence branching, size of SS relative to PS, and presence or absence of the PS, respectively. The number of orders of inflorescence branching excludes spikelet pairs. SS = sessile spikelet; PS = pedicellate spikelet.

*Andropogon abyssinicus*, 1, 1, SS > PS, present; *Andropogon burmanicus*, 2 or 3, 1, SS > PS, present; *Andropogon chinensis*, 1, 1, SS > PS, present; *Andropogon distachyos*, 1, 1, SS > PS, present; *Andropogon fastigiatus*, 0, NA, SS < PS, present; *Arthraxon hispidus*, 1 to 50, 1, SS > PS, absent; *Arthraxon lanceolatus*, 1 to 3, 1, SS > PS, present; *Arthraxon microphyllus*, 0 to 2, 1, SS > PS, present; *Arthraxon prionodes*, 1 to 7, 1, SS > PS, present; *Coix lacryma-jobi*, 0, 1, SS > PS, absent; *Chrysopogon gryllus*, many, 2 or more, SS = PS, present; *Chrysopogon orientalis*, many, 2 or more, SS > PS, present; *Chrysopogon serrulatus*, many, 2 or more, SS = PS, present; *Chrysopogon zizanioides*, many, 2 or more, SS = PS, present; *Cymbopogon flexuosus*, 1, 1, SS > PS, present; *Dichanthium sericeum*, 0 to 5, 1, SS > PS, present; *Dimeria ornithopoda*, 1, 1, absent, present; *Eremochloa ciliaris*, 0, NA, SS > PS, absent; *Eremochloa eriopoda*, 0, NA, SS > PS, absent; *Eriochrysis cayennensis*, many, 1, SS > PS, present; *Eriochrysis laxa*, many, 1, SS > PS, present; *Eriochrysis villosa*, many, 1, SS > PS, present; *Eulalia contorta*, 2 to 19, 1, SS = PS, present; *Eulalia siamensis*, 4 to 6, 1, SS = PS, present; *Eulaliopsis binata*, 1 to 3, 1, SS = PS, present; *Garnotia tenella*, many, 2 or more, SS = PS, present; *Germainia capitata*, 0, NA, SS < PS, present; *Heteropogon contortus*, 0, NA, SS < PS, present; *Heteropogon triticeus*, 0, NA, SS < PS, present; *Hemisorghum mekongense*, many, 2 or more, SS > PS, present; *Hyparrhenia diplandra*, 1, 1, SS = PS, present; *Hyparrhenia rufa*, 1, 1, SS = PS, present; *Kerriochloa siamensis*, 0, NA, SS > PS, present; *Mnesithea helferi*, 0, NA, SS > PS, rudimentary; *Parahyparrhenia siamensis*, 0 or 1, NA or 1, SS > PS, present; *Pogonatherum panicum*, 0, NA, SS = PS, present; *Polytoca digitata*, 0, NA, SS < PS, present; *Pseudosorghum fasciculare*, many, 2 or more, SS > PS, present; *Schizachyrium brevifolium*, 0, NA, SS > PS, rudimentary; *Schizachyrium exile*, 0, NA, SS > PS, rudimentary; *Schizachyrium imberbe*, 0, NA, SS > PS, present; *Schizachyrium sanguineum*, 0, NA, SS > PS, present; *Schizachyrium scoparium*, 0, NA, SS > PS, rudimentary or present; *Schizachyrium spicatum*, 0, NA, SS > PS, rudimentary; *Schizachyrium tenerum*, 0, NA, SS = PS, present; *Sorghum bicolor*, many, 2 or more, SS = PS, present; *Themeda arguens*, 0, NA, SS > PS, present; *Themeda arundinacea*, 0, NA, SS ≤ PS, present; *Themeda triandra*, 0, NA, SS = PS, present; *Themeda villosa*, 0, NA, SS = PS, present; *Zea mays*, 0, NA, SS > PS, absent.